EUROPEAN MEDICINES AGENCY
SCIENCE MEDICINES HEALTH

11 November 2021
EMA/CHMP/446813/2021
Committee for Medicinal Products for Human Use (CHMP)

Assessment report

Nouryant

International non-proprietary name: istradefylline

Procedure No. EMEA/H/C/005308/0000

Note

Assessment report as adopted by the CHMP with all information of a commercially confidential nature deleted.

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List of abbreviations

ADL	Activities of Daily Living
ADR	Adverse Drug Reaction
AEs	Adverse Events
AEoSIs	Adverse Events of Special Interest
ALT	Alanine Aminotransferase
ANOVA	Analysis Of Variance
ANCOVA	Analysis of Covariance
AST	Aspartate Aminotransferase
$AUC_{0\text{-}8h}$	Area Under the plasma Concentration versus time curve from 0 to 8 hours
$AUC_{0\text{-}24h}$	Area Under the plasma Concentration versus time curve from 0 to 24 hours
$AUC_{0\text{-}72h}$	Area Under the plasma Concentration versus time curve from 0 to 72 hours
$AUC_{0\text{-}\infty}$	Area Under the plasma Concentration versus time curve from 0 to infinity
$AUC_{0\text{-}\tau,ss}$	Area Under the dosing interval Concentration versus time curve at steady state.
AUEC	Area Under the Effect Curve
BCRP	Breast Cancer Resistance Protein
BMI	Body Mass Index
CK	Creatinin Kinase
CGI-I	Clinical Global Impression – Improvement scale
CGI-S	Clinical Global Impression - Severity scale
CI	Confidence Intervals
CL	Clearance
CL/F	Oral Clearance
C_{max}	Maximum Concentration observed
CNS	Central Nervous System
COMT	Catechol-O-Methyl Transferase
CRF	Case Report Form
QC	Quality Control
CV	Coefficient of Variation
DB	Double-Blind
DDI	Drug-Drug Interactions
DP-PR	Disease Progression/Placebo Response
DRF	Dose Range Finding
EAIR	Exposure-Adjusted Incidence Rates
EC_{50}	Half maximal effective concentration
EC_{75}	75% effective concentration
ED_{50}	Median Effective Dose
EFF	Efficacy Evaluable Population

EE	Efficacy Evaluable
EFD	Embryo-Foetal Development
Emax	Maximum effect
EPAR	European Public Assessment Report
E-R	Exposure-Response
ERA	Environmental Risk Assessment
FAS	Full Analysis Set
fss	fraction at steady state
6-OHDA	6-Hydroxydopamine
IC_{50}	Half Maximal Inhibitory Concentration
HPLC-UV	High-Performance Liquid Chromatography combined with UV detection
H&Y	Modified Hoehn and Yahr Scale
IRT	Interactive Response Technology
ISR	Incurred Sample Reanalysis
IPSCs	Inhibitory Post-Synaptic Currents
ITT	Intended To Treat
GD	Gestational Day
GLP	Good Laboratory Practices
GP	Globus Pallidus
LBM	Lean Body Mass
L/C	Levodopa/Carbidopa
LC-MS	HPLC combined with single quadrupole
LC/MS-MS	HPLC combined with tandem mass spectrometric detection
LLOQ	Lower Limit of Quantification
LOCF	Last Observation Carried Forward
LSM	Least-Squares Mean
M1	4'-O-demethyl istradefylline metabolites
M3	3’, 4’-O-didemethyl istradefylline
M4	4’-O-demethyl istradefylline conjugated to sulfate (sulfate conjugate of M1)
M5	4’-O-demethyl istradefylline conjugated to glucuronic acid
M8	1-β-hydroxylated istradefylline
M9	4’-O-demethyl-hydrogenated istradefylline
M10	3’, 4’-O-didemethyl-hydrogenated istradefylline
M11	4’-O-demethyl-hydrogenated istradefylline glucuronide
M12	3’, 4’-O-didemethyl-hydrogenated istradefylline monoglucuronide
M13	1-deethyl-3’, 4’-O-didemethyl-hydrogenated istradefylline
M14	1-β-hydroxylated-3’, 4’-O-didemethyl-hydrogenated istradefylline
M15	1-β-hydroxylated-3’, 4’-O-didemethyl-hydrogenated istradefylline monosulfate
M16	1-deethyl-4’-O-demethyl istradefylline sulfate

M17	1-β-carboxylated istradefylline
M18	3’, 4’-O-didemethyl-1-β-carboxylated-hydrogenated istradefylline
M19	3’, 4’-O-didemethyl-hydrogenated istradefylline monosulfate
MAO-B	Monoamine Oxidase B
MCID	Minimal Clinically Important Difference
MMRM	Mixed-model repeated measures
MMSE	Mini-Mental State Examination
MoCA	Montreal Cognitive Assessment
MPTP	1-Methyl-4-Phenyl-1,2,3,6-TetrahydroPyridine
mRNA	messenger RNA
MSN	Medium Spiny Neurons
NECA	5'-N-ethylcarboxamidoadenosine
NOAELs	No Observed Adverse Effect level(s)
OL	Open-Label
PBT	Persistence, Bioaccumulation, Toxicity
PCS	Potentially Clinically Significant
PD	Parkinson’s Disease; Pharmacodynamics
PET	Positron Emission Tomography
PK	Pharmacokinetics
PGI-I	Patient Global Impression - Improvement
P-gp	P-glycoprotein
PP	Per protocol
PQD	Parkinson’s Disease Questionnaire
PopPK	Population PK
QUIP-RS	Questionnaire for Impulsive-Compulsive Disorders in Parkinson's Disease - Rating Scale
RO	Receptor Occupancy
SA	Scientific Advice
SAE	Serious Adverse Events
SAG-N	Scientific Advisory Group Neurology
SAP	Statistical Analysis Plan
SB	Single-Blind
SD	Standard Deviation
SF-36	Study 36-Item Short Form
SNc	Substantia Nigra pars compacta
SOC	System Organ Class
TEAE	Treatment emergent adverse events
t_{max}	Time to reach maximum concentration
$t_{1/2}$	Term half-life

TK	Toxicokinetics
ULN	Upper Limit of Normal
UKPDS	United Kingdom Parkinson’s Disease Society
UPDRS	Unified Parkinson's Disease Rating Scale
Vd	Volume of distribution
Vdss	Volume of distribution steady-state
VPC	Visual Predictive Checks
VAS	Visual Analog Scale

1. Background information on the procedure

1.1. Submission of the dossier

The applicant Kyowa Kirin Holdings B.V. submitted on 25 November 2019 an application for marketing authorisation to the European Medicines Agency (EMA) for Nouryant, through the centralised procedure falling within the Article 3(1) and point 3 of Annex of Regulation (EC) No 726/2004. The eligibility to the centralised procedure was agreed upon by the EMA/CHMP on 28 February 2019.

The applicant applied for the following indication:

> Istradefylline is indicated in adults as an adjunctive treatment to levodopa-based regimens in patients with Parkinson's disease (PD) experiencing "OFF" time

The legal basis for this application refers to:

Article 8.3 of Directive 2001/83/EC - complete and independent application

The application submitted is composed of administrative information, complete quality data, non-clinical and clinical data based on applicants' own tests and studies and/or bibliographic literature substituting/supporting certain test(s) or study(ies).

Information on Paediatric requirements

Pursuant to Article 7 of Regulation (EC) No 1901/2006, the application included an EMA Decision(s) P/0270/2019 on the granting of a (product-specific) waiver.

Information relating to orphan market exclusivity

Similarity

Pursuant to Article 8 of Regulation (EC) No. 141/2000 and Article 3 of Commission Regulation (EC) No 847/2000, the applicant did not submit a critical report addressing the possible similarity with authorised orphan medicinal products because there is no authorised orphan medicinal product for a condition related to the proposed indication.

Applicant's request(s) for consideration

New active Substance status

The applicant requested the active substance istradefylline contained in the above medicinal product to be considered as a new active substance, as the applicant claims that it is not a constituent of a medicinal product previously authorised within the European Union.

Scientific advice

The applicant received the following Scientific Advice (SA) on the development relevant for the indication subject to the present application:

Date	Reference	SAWP Coordinators
23 September 2003	EMEA/H/SA/449/1/2003/III	Prof. Fernando de Andres Trellés Dr Barbara van Zwieten-Boot

12 September 2005	EMEA/H/SA/449/2/2005/I	Dr Clemens Mittmann Dr Stefano Vella

The EMEA/H/SA/449/1/2003/III pertained to non-clinical and clinical aspects:

- CHMP agreed with use of Hauser diary to score presence/absence of dyskinesia. CHMP considered using a comparator that is directly acting e.g Dopamine agonist might be more beneficial but agreed that Entacapone as comparator in proposed Phase III study could be sufficient.
- Recommendations made in SA by CHMP which the applicant did not fully adhere to include controlling for multiplicity of secondary endpoints and to study the test drug in patients receiving L-dopa+ only

The EMEA/H/SA/449/2/2005/I SA procedure pertained to the following quality aspects:

- Agreement in nominating NAUE and DMCA as Starting materials for the manufacturer of istradefylline
- Agreement that these data are suitable to support multiple suppliers
- Agreement that the data supplied are sufficient to confirm that the drug substance exists as only one polymorphic form during the manufacturing processes for drug product and stability testing?
- Agreement that the disintegration test for istradefylline tablets can be replaced by a dissolution test as a pivotal parameter in the finished product specification.
- Agreement with the company's decision making process for developing a non-pharmacopoeial dissolution method to ensure batch to batch consistency

1.2. Steps taken for the assessment of the product

The Rapporteur and Co-Rapporteur appointed by the CHMP were:

Rapporteur: Jayne Crowe Co-Rapporteur: Armando Genazzani

The application was received by the EMA on	25 November 2019
The procedure started on	2 January 2020
The Rapporteur's first Assessment Report was circulated to all CHMP members on	23 March 2020
The Co-Rapporteur's first Assessment Report was circulated to all CHMP members on	31 March 2020
The PRAC Rapporteur's first Assessment Report was circulated to all PRAC members on	2 April 2020
The CHMP agreed on the consolidated List of Questions to be sent to the applicant during the meeting on	30 April 2020
The following GCP inspection was requested by the CHMP and its outcome taken into consideration as part of the Quality/Safety/Efficacy assessment of the product: - A GCP inspection at one clinical investigator site in Poland and at the Sponsor site in US between March and July 2020. The outcome of the inspection carried out was issued on	14 October 2020
The applicant submitted the responses to the CHMP consolidated List of Questions on	20 December 2020
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of	01 February 2021

Questions to all CHMP members on	
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	11 February 2021
The CHMP agreed on a list of outstanding issues in writing and/or in an oral explanation to be sent to the applicant on	25 February 2021
The applicant submitted the responses to the CHMP List of Outstanding Issues on	21 May 2021
The Rapporteurs circulated the Joint Assessment Report on the responses to the List of Outstanding Issues to all CHMP members on	09 June 2021
The outstanding issues were addressed by the applicant during an oral explanation before the CHMP during the meeting on	23 June 2021
The CHMP, in the light of the overall data submitted and the scientific discussion within the Committee, issued a negative opinion for granting a marketing authorisation to Nouryant on	22 July 2021

1.3. Steps taken for the re-examination procedure

The Rapporteur and Co-Rapporteur appointed by the CHMP were:

Rapporteur: Paula van Hennik Co-Rapporteur: Maria Concepcion Prieto Yerro

The applicant submitted written notice to the EMA, to request a re-examination of Istradefylline KKHBV CHMP opinion of 22 July 2021, on	30 July 2021
The CHMP appointed Paula van Hennik as Rapporteur and Maria Concepcion Prieto Yerro as Co-Rapporteur on	16 September 2021
The applicant submitted the detailed grounds for the re-examination (Appendix X of Final Opinion) on	13 September 2021
The re-examination procedure started on	14 September 2021
The Rapporteur's re-examination assessment report was circulated to all CHMP members on	13 October 2021
The Co-Rapporteur's assessment report was circulated to all CHMP members on	13 October 2021
The Rapporteurs circulated the Joint Assessment Report on the detailed grounds for re-examination to all CHMP members on	5 November 2021
SAG/Expert group were convened to address questions raised by the CHMP on The CHMP considered the views of the SAG as presented in the minutes of this meeting	4 November 2021
The detailed grounds for re-examination were addressed by the applicant during an oral explanation before the CHMP on	9 November 2021
The CHMP, in the light of the scientific data available and the scientific discussion within the Committee, re-examined its initial opinion and in its final opinion concluded that the application did not satisfy the criteria for authorisation and did not recommend the granting of the marketing authorisation	11 November 2021

2. Scientific discussion

2.1. Problem statement

2.1.1. Disease

Parkinson's disease (PD) is a progressive, debilitating movement disorder characterized by bradykinesia, rigidity, and resting tremor that affects 1.2 million people in Europe (European Brain Council, 2019).

2.1.2. Epidemiology

Estimates of the number of people in Europe (Western, Eastern, and Central) with PD range from 1.2 million (Andlin-Sobocki, 2005) to 1.4 million (GBD 2016 Parkinson's Disease Collaborators, 2018). In the US, the estimated number of persons with PD varies from one half million (U.S. Department of Health & Human Services. Food and Drug Administration. NIH RePORT) to 1 million (Parkinson's Foundation), while in Japan approximately 145,000 people have PD (Fujimoto, 2011). The differences in prevalence may be attributed to differences in the sizes of the populations (Dorsey, 2007).

2.1.3. Clinical presentation

The hallmark symptoms of bradykinesia, rigidity, tremor, and loss of postural reflexes. In the early stages of PD, patients usually experience substantial symptom relief from levodopa (Ahlskog, 2001). Despite its initial efficacy, levodopa's therapeutic window narrows over time (Jankovic, 2005). That is, the duration of benefit from a dose of levodopa ("ON time") becomes shorter and PD symptoms return before the next scheduled dose ("wearing-off"). There are periods of time when, despite measurable plasma concentrations, levodopa does not control PD symptoms ("OFF time") and these periods become increasingly longer as PD progresses. These motor fluctuations appear in about 50% of levodopa-treated patients who have received the drug for more than 5 years (Hickey, 2011). Another complication of levodopa therapy is the development of dyskinesia, which usually occurs within 3 to 6 years after the initiation of treatment and affects 30% to 80% of patients (DeMaagd, 2015).

2.1.4. Management

Since the pathobiology of PD is characterized by a loss of dopamine in the basal ganglia, current medical therapy focuses primarily on restoring dopamine in those parts of the brain. Levodopa remains the "gold standard" for therapy that increases dopamine concentrations in the brain because levodopa can cross the blood-brain barrier and be transformed into dopamine.

The same categories of available symptomatic treatments for PD are used globally: levodopa (always in combination with a DOPA-decarboxylase inhibitor), dopamine agonists, monoamine oxidase B (MAO-B) inhibitors, and catechol-O-methyl transferase (COMT)-inhibitors which extend the peripheral half-life of levodopa. Amantadine and anticholinergic agents are also used. There may be regional differences in the order in which these medications are prescribed as adjunctive treatment to levodopa. Nevertheless, across all regions, levodopa or a dopamine agonist are generally recommended for first-line treatment of patients with PD, with dopamine agonists avoided in elderly patients. However, there may be variations in concomitant anti-parkinson medications between Europe, the US, and Japan due to differences between regions in availability of medications. For example, several anti-parkinson medications available in Europe (Rosa, 2010) are not approved in the US: the dopamine agonists (budipine, dihydroergocryptine, lisuride), and the anticholinergics (bornaprine, dexetimide).

Efficacy of available treatments varies from patient-to-patient and over time. There are many different types of dopaminergic therapies that have been developed to try to treat OFF time (e.g., dopamine agonists, COMT inhibitors, monoamine oxidase B [MAO-B] inhibitors); however, treatment is complicated by an increased risk of dyskinesia and a variety of side effects, including impulse control disorders and sleep disturbances, which may limit benefits or preclude continued use of these medications. Thus, despite available medical therapies, patients continue to suffer potentially disabling OFF episodes. Consequently, there is a continuing need for additional agents that are effective for the levodopa-treated patient (producing less OFF time and better symptom control) without intolerable side effects. The clinical heterogeneity of PD signs and symptoms and in the individual levodopa requirements reinforces the need for additional adjunctive treatment options (Lewis, 2005).

About the product

Istradefylline (also known as KW-6002) is an adenosine A_{2A} receptor antagonist, which has a xanthine derivative structure and which competitively inhibits adenosine binding to the A_{2A} receptor. Istradefylline is a potent, selective, and competitive antagonist of the A_{2A} receptor and does not demonstrate any significant affinity for other neurotransmitter receptors, including dopamine receptors (D1, D2, D3, D4, and D5). In addition, istradefylline did not demonstrate inhibition of MAO-A, MAO-B, or COMT (Saki, 2013). Since istradefylline does not affect dopamine concentrations or dopamine receptor activation, it may be considered a nondopaminergic agent.

The highest concentration of A_{2A} receptors in the brain is found in the indirect pathway of the basal ganglia. These receptors are specifically and highly expressed on the medium spiny neurons of the striatum, which also express D2 receptors. Dopamine activation of the D2 receptor suppresses the indirect pathway. With loss of dopamine in PD, increased excitability of the indirect pathway results. Increased excitability of the indirect pathway can also be mediated by adenosine activation of the A_{2A} receptors in the striatum and in the globus pallidus, i.e., A_{2A} receptor-mediated dual modulation (Mori, 2003). Blockade of A2A receptors by istradefylline decreases the excessive activation of the indirect pathway in PD and restores the balance in the basal ganglia-thalamocortical circuit. The blockade of A_{2A} receptors occurs without affecting the direct pathway (Mori, 2014) and does not affect dopamine concentrations or dopamine receptor activation (Mori, 2015). Thus, istradefylline provides an alternative, non-dopaminergic approach to symptomatic relief of PD.

The target indication for istradefylline is as an adjunctive treatment to levodopa-based regimens in adult patients with PD experiencing "OFF" time. The recommended starting dose of istradefylline is 20 mg administered orally, once a day. The dosage may however be initiated at, or increased to, a maximum of 40 mg once daily, based on clinical need and individual tolerability. Istradefylline can be taken with or without food. The formulation proposed is an immediate-release tablet of istradefylline.

Type of Application and aspects on development

The applicant has submitted 57 clinical studies, Phase 1-3, conducted in healthy volunteers and patients with PD. Of these, there are 2 studies submitted in patients with Restless Leg Syndrome and 2 in patients with Major Depressive Disorder which will not be considered further in this report. The remaining studies are discussed in the relevant sections.

It is notable that clinical development began over 20 years ago and of the pivotal trials, four were conducted between 2002 and 2005, the next two in Japan between 2007 and 2011 and the most recent pivotal trial completed in 2016.

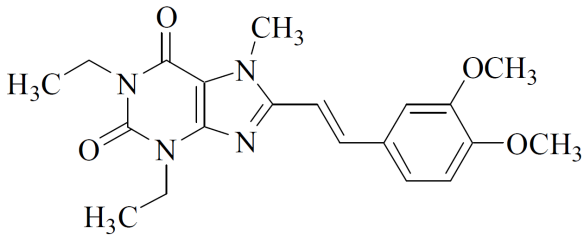
2.2. Quality aspects

2.2.1. Introduction

The information to support the quality of the chemical drug substance istradefylline and the incorporation of this drug substance into 20 mg and 40 mg film-coated tablets is contained in the dossier provided by the applicant.

2.2.2. Active Substance

The drug substance is an aqueously insoluble light yellow-green crystalline material that is manufactured in one stable polymorphic form.



Molecular formula: $C_{20}H_{24}N_4O_4$
Relative molecular mass: 384.43

It is aqueously insoluble across the physiological pH range. It is non-hygroscopic. It has no chiral centres but has a trans configuration across a double bond, where the cis configuration structure is a noted impurity.

The drug substance manufacturing site has an appropriate declaration of GMP compliance that has been provided. The manufacturing process is sufficiently described. The overall control of the manufacturing process encompasses the control over the starting materials, the in-process controls, the intermediate specifications, the process parameters and their PARs, and the identification and control of the CPPs for the process. The proposed starting materials are acceptable based on the supporting data provided, and the starting material test method capability has been confirmed. The evaluation of the risk of the formation of nitrosamines has been addressed.

A broad range of process parameters and associated ranges have been investigated to understand the drug substance manufacturing process and the required control over the process. This has enabled the establishment of routine production process parameter controls (NORs) and associated PARs. Sufficient information has been provided on the experimental programme used to investigate the process parameters as well as how the PARs were determined.

The methology for establishing whether a parameter is a CPP has been satisfactorily explained. The process has sufficient in-process and intermediate controls to facilitate the requisite drug substance quality, and the suitability of the analytical methods used in these controls has been verified.

The drug substance structure has been characterised. This characterisation allows the spectral differentiation between the drug substance trans and impurity cis configurations. The potential for genotoxic impurities in istradefylline prepared by the current route of manufacture has been evaluated through a comprehensive risk assessment. Four genotoxic impurities have been shown to be adequately purged during the synthesis. Three of these impurities will have a non-routine control in the drug substance specification.

The drug substance specification contains the necessary tests for a drug substance and the specification limits can be accepted. The analytical methods have been described and they have been appropriately validated The drug substance test method reference standards are appropriately certified.

The container closure system is suitably described. The drug substance is stored and transported in closed, double Low-Density Polyethylene (LDPE) bags within a drum with a secure fitting lid. The polyethylene bags meet the requirements for plastics intended to come in contact with food.

The drug substance stability studies were conducted in accordance with ICH Q1A (R2), Stability Testing of New Drug Substances and Products, using the general-case long-term storage conditions of 25°C±2°C at 60%±5% relative humidity (RH), and 40°C±2°C at 75%±5% RH for accelerated stability storage conditions with 6 months data available. In addition, the experimental stress degradation, photostability, and storage container compatibility studies were also conducted and described in this section. The stability demonstrates good stability of the drug substance and supports the proposed retest.

2.2.3. Finished Medicinal Product

The drug product, Istradefylline 20 mg and 40 mg film-coated tablets, contain 20 mg and 40 mg of istradefylline drug substance, respectively. The tablets are comprised of a commonly used ingredients for this dosage form and the two strengths are proportional in composition. They are supplied in two different presentations: high density polyethylene (HDPE) bottle sealed with an aluminium foil liner and closure, and blisters.

The drug product development looked at the impact of formulation variations on the product CQAs in order to arrive at the optimum formulation. The manufacturing process was investigated in order to determine the CPPs and the associated necessary controls. This development work is informative and helps understand the controls necessary in the formulation and manufacturing process. A dissolution method was developed that uses a medium with surfactant to facilitate the dissolution of the aqueously insoluble drug substance while still providing some discriminatory ability. The dissolution method and specification have been appropriately justified. An Establishment Licence issued by Health Canada based on a recent inspection supports that the site operates under an acceptable GMP status.

The drug product is manufactured at a commercial batch size and the manufacturing process parameters are controlled based on the outcome of the development work. Process validation has been completed and the manufacturer has experience of commercial manufacture of the product for markets outside the EU. The excipients are all controlled by suitable specifications and pharmacopoeial or standard methods.

The drug product specification is acceptable.

The analytical procedures to control the drug product are adequately described and are suitably validated. Batch analysis data shows the ability of the manufacturer to consistently make product to the required testing specification. There are no other degradation impurities other than those identified in the drug substance. A major objection that had been raised in relation to the provision of the risk evaluation concerning the potential presence of nitrosamines in the drug product has been resolved. A suitable reference standard is used in the testing of the product.

The container closure systems (blister packages and HDPE containers) have been sufficiently described, and the child resistant properties of the HDPE container have been confirmed. The istradefylline tablet stability studies were conducted in accordance with ICH Q1A (R2), Stability Testing of New Drug Substances and Products, using the general-case, long-term storage conditions of 30°C±2°C at 65%±5% RH for the registration batches, process validation batches, and commercial batches. The accelerated storage conditions were set at 40°C±2°C and 75%±5% RH. In addition to the long-term and accelerated storage condition stability studies, ICH photostability studies were conducted according to ICH Q1B,

Photostability Testing of New Drug Substances and Products. Data for the long-term stability studies and the accelerated stability studies conducted on registration batches and process validation batches are available. Evaluation for Stability Data. Istradefylline tablets are packaged into the high density polyethylene (HDPE) bottles or blisters with no special storage conditions required.

2.2.4. Conclusions on chemical, pharmaceutical and biological aspects

The documentation provided to support the quality aspects of the application is of an acceptable standard. Sufficient information is provided to be able to evaluate the manufacturers' understanding and proposed control of the drug substance and drug product manufacturing processes. A major objection raised in relation to the absence of a risk evaluation into the potential formation and consequential control of nitrosamines in the drug substance and drug product has been resolved. The other concerns raised about the manufacturing and testing of the drug substance and drug product have been resolved.

2.2.5. Recommendation(s) for future quality development

Not applicable

2.3. Non-clinical aspects

2.3.1. Pharmacology

Primary pharmacodynamic studies

The applicant has demonstrated the selectivity of istradefylline for A_{2A} receptors over other adenosine receptor subtypes *in vitro*, with a binding affinity of istradefylline for A_{2A} receptors in the low nanomolar range (Ki= 12.4 nmol/L) in recombinant human receptors which is 10-fold greater that the affinity for human A_{2B} receptors (Ki = 150nmol/L). A significantly greater difference in affinity for A_{2A} over human A_1 (IC_{50} >1000nM) and A_3 receptors (IC_{50} >10,000) is also reported. There are some species differences in affinity of istradefylline for A_{2A} receptors (A_{2A} Ki=2.2, 8.18, 65, 11 nmol/L in mouse, rat, dog, and marmoset respectively). In dogs istradefylline presented the lowest affinity for A_{2A} receptors (Ki = 65 nmol/L) and the lowest selectivity for A_{2A} over A_1 receptor (3.4-fold in dog, vs >80-fold in human, and 14-fold in rat). The applicant justified the choice of dog as a nonclinical species for toxicity testing on the basis that monkeys presented a low absorption of istradefylline and there is precedent for the use of dogs in these types of studies. The kinetic parameters of istradefylline binding were analysed and the association and dissociation kinetic rate constants indicate that istradefylline binding to human and rat A_{2A} receptors is saturable and reversible. The specificity of istradefylline was also demonstrated *in vitro*, with weak affinity for other neurotransmitter receptor types including dopamine, serotonin and noradrenaline receptor subtypes.

Regarding mechanism of action, istradefylline did not affect basal cyclic AMP (cAMP) levels in PC-12 cells expressing adenosine A_{2A} receptors, indicating that it is not an agonist for these receptors. However, istradefylline competitively inhibited intracellular cAMP accumulation induced by a selective A_{2A} receptor agonist (CGS21680) in PC-12 cells expressing A_{2A} receptors (K_B = 0.74 ± 0.23 nmol/L). *In vivo*, carbon-14 labelled istradefylline demonstrated binding in the striatum, nucleus accumbens and olfactory tubercle of rats, a distribution pattern that corresponds with known A_{2A} receptor localization in the medium spiny neurons (MSN) of the indirect striatal output pathway. In a study to assess the effect of istradefylline on GABAergic synaptic transmission in the globus pallidus (GP), istradefylline inhibited a CGS21680-induced increase in the amplitude of $GABA_A$ receptor-mediated inhibitory post-synaptic currents (IPSCs) in patch clamp recordings of GP neurons in rat brain slices. These data support the rationale for using A_{2A} blockade to reduce GABAergic inhibitory outputs to the GP. Furthermore, in support of the proposed utility of A_{2A}

blockade in PD, basal GABA release was shown in a microdialysis study to be increased in the 6-hydroxydopamine (6-OHDA)-lesioned rats, suggesting hyperactivity in the striatopallidal pathway of this rat PD model. Istradefylline administration (1mg/kg po) attenuated the increased GABA release by approximately 20% in 6-OHDA-lesioned rats, suggesting an attenuation of hyperactivity in the striatopallidal indirect pathway of lesioned rats. Oral istradefylline (2.5mg/kg) was also shown to increase locomotor activity in the open field in $D_2R^{-/-}$ mice, who have a locomotor phenotype analogous to PD. These data indicate that A_{2A} receptor antagonism increased locomotor activity independent of D_2 receptors, despite the co-localization of adenosine A_{2A} receptors and dopamine D_2 receptors are on the striatopallidal MSNs.

Proof-of-concept studies conducted in rodent PD models demonstrate that istradefylline antagonizes CGS21680-induced catalepsy (ED_{50} = 0.05 mg/kg) and hypomobility in mice. Also istradefylline antagonized haloperidol-induced catalepsy (ED_{50} =0.03 mg/kg, 0.06 mg/kg in mice and rats respectively) more potently than bromocriptine (dopamine agonist) and L-DOPA in mice and rats. It was noted that there was no evidence of tolerance to the inhibitory activity in this haloperidol-induced catalepsy model, with istradefylline administered for up to 14-days. However, inhibitory activity beyond 14 days was not examined in any proof of concept study and notably hyperactivity findings in the rat repeat-dose toxicology studies resolve at approximately day 14 of istradefylline administration. Istradefylline was shown to reverse motor disability in MPTP treated marmosets in a dose-dependent manner for 21 days (Kanda, 1998, study #98-216), but evidence of a durability of effect of istradefylline beyond 21 days in non-clinical species is not available. However, the durability of effect of istradefylline is addressed in the clinical efficacy assessment and therefore, further non-clinical data will not be requested.

The dopaminergic neurotoxin 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) provides the basis for an additional animal model of PD, by inducing severe degeneration of the dopaminergic nerve terminals (>80%), leading to akinesia. In the MPTP mouse model of PD, istradefylline dose-dependently (0.16–2.5 mg/kg) antagonized a marked reduction in locomotor activity under loss of dopaminergic input from the SNc. Furthermore, unilateral intracerebroventricular injection with 6-OHDa in rats causes loss of dopaminergic neurons in the SNc, providing a model of hemiparkinsonism. Administration of dopamine receptor agonists such as apomorphine in this model causes contralateral turning behaviour in these animals. Istradefylline (1-10 mg/kg po) significantly potentiated and prolonged the effects of apomorphine in a dose-dependent manner but produced only weak and short-lasting contralateral turning when administered alone.

In MPTP-treated common marmosets, istradefylline treatment improved motor disability (bradykinesia, rigidity, abnormal posture of limbs and trunk) at all dose levels (0.5 to 100mg/kg), while istradefylline-induced increased locomotor activity at doses ≥ 5 mg/kg, although a clear dose-response relationship is not apparent for either measure. The effect of istradefylline on motor disability at a dose of 10 mg/kg was demonstrated to be significant between 10 minutes and at least 10 hours post-administration. Furthermore, comparing the effects of L-DOPA and istradefylline on locomotor activity in MPTP-treated and control marmosets indicates that istradefylline increased locomotor activity in MPTP-treated animals to the level of normal animals, while L-DOPA (10mg/kg) caused hyperactivity. Istradefylline (10 mg/kg) is also reported to enhance the effects of a suboptimal dose of Levodopa/ Carbidopa (L/C; levodopa: 2.5 mg/kg) on locomotor activity. However, the data presented indicate an increase in activity with respect to the vehicle group only, therefore the reported enhancement in the effect of suboptimal levodopa may not be statistically significant.

A blinded four-way crossover study was conducted to correlate pharmacodynamic activity with pharmacokinetic parameters. Istradefylline was dosed between 0.3 and 10mg/kg and a positive association between plasma concentrations of istradefylline and anti-parkinsonian activity in MPTP-treated common marmosets is reported. The applicant indicates that istradefylline dose-dependently

decreases total disability score, but each dose level is demonstrated to be significantly different from vehicle only, no significant differences between dose levels are demonstrated. For locomotor activity, the istradefylline-induced increase is only statistically significant in comparison to vehicle at the highest dose of 10mg/kg. Pharmacokinetic (PK) parameters all increased with increasing istradefylline dose from 0.3mg/kg to 10mg/kg in MPTP-treated marmosets. Time to reach maximum concentration (t_{max}) ranged from 1.5 to 5.2 h, maximum concentration observed (C_{max}) from 11 to 201.7 ng/ml, area under the plasma concentration versus time curve from 0 to 8 hours (AUC_{0-8h}) from 54.2 to 1185.5 ng.h/ml and area under the plasma concentration versus time curve from 0 to 72 hours (AUC_{0-72h}) from 84.3 to 2922.6 ng.h/ml.

In MPTP-lesioned marmosets, levodopa (10mg/kg) administered twice daily for 4 weeks induced mild dyskinesia. When these animals were subsequently dosed with istradefylline (10mg/kg) for 21 days, and a further single dose 1 week after chronic dosing (day 28), little or no dyskinesia is reported. These data indicate that istradefylline does not induce dyskinesia in this L-DOPA-primed MPTP-lesioned common marmoset model at doses previously demonstrated to ameliorate parkinsonian symptoms in MPTP-treated marmosets. Sub-optimal L-DOPA (2.5mg/kg) also induced mild dyskinesia in this model, although significantly less than the initial dyskinetic response, and co-administration of istradefylline for 21 days reduced the amplitude of these involuntary movements. Istradefylline co-administered with suboptimal L-DOPA 1 week after chronic dosing (day 28) also induced milder dyskinesia that the effect of suboptimal L-DOPA alone. However, the dyskinetic response to the optimal dose of L-DOPA (10mg/kg) was unchanged by chronic treatment with istradefylline. Therefore, while it has been shown that istradefylline does not induce dyskinesia in this model, it also does not ameliorate L-DOPA-induced dyskinesia when L-DOPA is dosed optimally to ameliorate parkinsonian symptoms.

Istradefylline at doses ranging from 60 to 90 mg/kg, also reversed motor disability in MPTP-treated cynomolgus monkeys. High doses of oral istradefylline were required in this species due to low absorption of istradefylline in cynomolgus monkeys. The magnitude of the effect obtained with 90 mg/kg of istradefylline was comparable to that observed with an optimal dose of L-DOPA (levodopa: 50 mg). Unlike L-DOPA, however, istradefylline did not produce dyskinesia. When istradefylline (60 to 90 mg/kg) was added to 50 mg of L-DOPA, dyskinesia was not further aggravated. Hence, in agreement with the MPTP-treated common marmosets' data, istradefylline improved motor disability without inducing dyskinesia in the MPTP-treated cynomolgus monkey.

A cis isomer (KF23546) and dimer (KF66275) of istradefylline have been identified as impurities in drug substance. The affinity of each for rat A_{2A} receptor is weaker than istradefylline, with $IC_{50s} \geq 1\mu M$ for both. The A_{2A} antagonist activity was also assessed at 30 and 100mg/kg doses of each *in vivo* reserpine-induced catalepsy model and the % inhibition of the catalepsy was significantly lower than that of 1mg/kg istradefylline. As KF23546 and KF66275 have been detected at < 0.1% in the drug substance, they are unlikely to significantly contribute to the pharmacological activity of istradefylline.

Metabolites observed in the plasma of rats, dogs, and humans were 4'-O-demethyl istradefylline (M1), 1-β-hydroxylated istradefylline (M8), 4'-O-demethyl istradefylline conjugated to sulfate (M4), and 4'-O-demethyl istradefylline conjugated to glucuronic acid (M5). The applicant provides a comparison of the affinity of Istradefylline and the metabolites M1, 3', 4'-O-didemethyl istradefylline (M3) and M4 for rat A_{2A} receptors. However, an assessment of the metabolite profile of istradefylline in mouse, rat, dog and human plasma using carbon-14 labelled istradefylline are included in the PK section. These data indicate a plasma content of M5>M1>M4 in rats, while M3 is a major metabolite in urine and faeces but not in plasma. Hence, it is unclear why these particular metabolites were chosen for binding affinity comparison. Istradefylline, M1 and M4 had similar affinities for rat A_{2A} receptor with M4> M1 > istradefylline (Ki = 4.8, 6.35, 8.18 nmol/L respectively), while M3 had approximately 3-fold lower affinity. Istradefylline and M1 also had the same affinity for human A_{2A} receptors (Ki=12.4nmol/L), but no other metabolite binding affinities are reported in human A_{2A} receptors. The plasma content of M1 appears to

be low in humans and the binding affinities of M5 and M8 for human A_{2A} receptors may be more relevant. Therefore, the metabolic profile of istradefylline in rats and dogs was only qualitatively but non quantitatively similar to that observed in humans. According to the applicant, none of M4, M5 and M8 are pharmacologically active. Unfortunately, this is not supported from a non-clinical PD point of view since M4 presented an even greater affinity for rat A_{2A} receptor compared to istradefylline, while the affinity of M5 and M8 for the A_{2A} receptor hasn't been tested in any species. The applicant also compared the activity of istradefylline, M1 and M3 in two *in vivo* mouse models, a clonidine-induced aggressive behaviour model and a haloperidol-induced catalepsy model. M1 demonstrated activity in both models while M3 was active in neither model up to a dose of 10mg/kg. Indeed, M1 had similar potency (ED_{50} = 0.08mg/kg) to istradefylline (ED_{50}=0.03mg/kg) in the haloperidol-induced catalepsy model, although it was considerably less potent in the clonidine-induced aggression model (ED_{50}=10mg/kg) than istradefylline (ED_{50}=0.63mg/kg). These data provide an incomplete picture of the potential contribution of the metabolites to the PD. While the applicant has provided data to show a similar binding affinity of M1 and istradefylline for human A_{2A} receptors and a similar potency of M1 and istradefylline in the haloperidol-induced catalepsy model in mice, it appears that the level of M1 present in human plasma is low and the binding affinity and activity of other more relevant metabolites is not reported. Given the metabolite profile data indicating a higher content of M5 and M8 in human plasma, and the binding affinity data indicating a higher affinity of M4 for the A_{2A} receptor in rats than istradefylline, the applicant was asked to discuss the binding affinities of M4, M5 and M8 in human adenosine receptors, their relative potency to istradefylline and potential contribution to the *in vivo* effect of istradefylline. The applicant clarified that although the binding affinities of the metabolites M4, M5 and M8 to human A_{2A} receptors are unknown, the contribution of these metabolites to the observed PD activity of istradefylline is considered likely to be low. This is based on reported exposure levels for each of the metabolites in question of < 10% of the exposure to the parent drug, following 14 days of istradefylline dosing in humans (Study 6002-US-016).

Secondary pharmacodynamic studies

Secondary PD studies focussed on the cardiovascular effects of istradefylline but studies evaluating central nervous system (CNS), inflammatory effects and potential platelet aggregation effects were also included. The applicant indicates istradefylline was found to be effect in rodent models of depression and restless-leg syndrome but there were no findings of note regarding the safety of istradefylline in these CNS studies.

The potential effect of istradefylline on the incidence of ventricular arrhythmia following coronary artery ligation and the potential effect on coronary vasodilatory reserve (as measured by reactive hyperaemic response) were investigated in dogs, but no significant treatment-related effects are reported and istradefylline was considered to pose minimal risks in terms of arrhythmia generation. The effects of istradefylline on 5'-N-ethylcarboxamidoadenosine (NECA; a nonselective adenosine receptor agonist)-induced hypotension and bradycardia were also examined in rats. Istradefylline (0.3, 1, 3, 10 mg/kg po) inhibited NECA-induced hypotension (mediated via A_{2A} receptors) but did not affect NECA-induced bradycardia (mediated via A_1 receptors), indicating that direct effects of istradefylline on the cardiovascular system occurred via a selective and potent adenosine A_{2A} antagonistic action in rats at doses up to 10 mg/kg.

An anti-inflammatory effect of istradefylline in a carageenan-induced inflammation model of unknown relevance is reported. No effect of istradefylline on either ADP- or collagen-induced platelet aggregation in rabbit platelet rich plasma is reported.

During the scientific advice procedure CHMP raised an issue regarding the lack of secondary pharmacology information and the need for an evaluation of the activity of istradefylline at peripheral

(skeletal muscle) A_{2A} receptors. Although this issue has not been directly addressed by the secondary pharmacology studies presented, a GLP-compliant study in anaesthetised dogs assessed femoral blood flow is included in the safety pharmacology and further non-clinical studies are unlikely to add value to the non-clinical safety assessment.

Safety pharmacology programme

CNS safety pharmacology studies in mice, including a good laboratory practices (GLP)-compliant study (#KHK23/952834), demonstrate increased spontaneous motor activity following ≥ 0.16mg/kg istradefylline, these increases in spontaneous locomotor activity were maximal at 30mg/kg and effects lasted at least 10 hours. The applicant includes another GLP-compliant study which indicates decreased locomotor activity in mice at 100 and 300mg/kg istradefylline and no significant effect at 30mg/kg (KHK24/952828). No explanation for this divergent result is provided. However, istradefylline-induce increased locomotor activity is also reported in naive common marmosets at single oral doses ≥3mg/kg, with maximal effect at 10 mg/kg and no further increase in activity at higher doses up to 300mg/kg. Istradefylline also restored locomotor activity in MPTP-treated marmosets to the level of normal animals, no hyperactivity is reported. Other reported CNS effects are limited to significantly shortened pentobarbital-Na-induced sleeping time at ≥10mg/kg and increased rectal temperatures at ≥ 0.3mg/kg istradefylline in mice. Istradefylline-induced increased in rectal temperatures also occurred in rats (≥1.0 mg/kg) and rabbits (300mg/kg).

CVS safety pharmacology studies included an *in vitro* hERG assay in HEK293 cells to assess the potential effects of istradefylline on ventricular repolarization and risk for QT prolongation (d-05-140). Istradefylline at concentrations up to 2 µmol/L (1.71 µmol/L, the maximum soluble concentration in this test condition), did not inhibit the hERG tail current. This maximal concentration is approximately 64 or 27-fold higher than plasma protein free fraction of the mean C_{max} achieved in humans receiving istradefylline at 20 or 40 mg/day, respectively. This study is not GLP compliant, the applicant has provided a justification for this on the basis that the study meets reliability, reproducibility and record keeping criteria outlined in the "Criteria for Reliability of Application Data (Article 43 of the Enforcement Regulations, Pharmaceutical Affairs Law)" of Japan, where the study was performed. The sponsor indicates that the lack of GLP-compliance does not impact on study endpoints. Oral administration of istradefylline at 0.3 to 10 mg/kg in conscious rats increased blood pressure and heart rate. These effects were interpreted to be associated with a dose-dependent increase in locomotor activity in these animals, the effect on blood pressure and heart rate were not dose-dependent. In the same study report, no effects on heart rate or electrocardiogram (ECG) were reported in conscious dogs administered istradefylline at doses of 25 – 200mg/kg, locomotor activity was not recorded in these dogs. The applicant acknowledges that istradefylline is less potent as an adenosine A_{2A} antagonist in dogs compared to humans and rats. Likewise, there is less selectivity in the dog for A_{2A} vs A_1 antagonism compared to humans. However, the evaluation of istradefylline's effects was considered valid as istradefylline concentrations achieved in the plasma dogs during cardiovascular safety assessment were above that required for inhibition of adenosine A_{2A} receptors, although it's expected that significant inhibition of A_1 receptors as well as A_{2A} receptors had been achieved in the dogs whereas significant inhibition of A_1 receptors in humans at the usual clinical doses of 20 mg/day or 40 mg/day is unlikely.

A GLP-compliant study was also conducted in conscious dogs (KYW011DG) in which arterial blood pressure, heart rate, and the Lead II electrocardiogram were unchanged following single-dose oral administration of istradefylline at doses of 8, 40, or 400 mg/kg. No clinically significant effects on the cardiovascular system, the respiratory system, general activity and behaviour, or body temperature are reported. The total C_{max} achieved in dogs at the highest dose of 400mg/kg resulted in exposure multiples relative to humans receiving the 20mg and 40mg daily doses of 9.2 and 3.9 respectively. The C_{max} for unbound istradefylline in dogs at 400mg/kg indicates somewhat higher exposure margins of 18.4 and

7.9 relative to the 20mg and 40mg clinical doses respectively, although istradefylline is approximately 5-fold less potent in dogs than in humans (Ki=65 nmol/L in dogs and 12.4 nmol/L in humans).

A reduction in femoral blood flow and a concurrent increase in calculated femoral resistance was consistently observed in a GLP-compliant study in anaesthetised dogs following intraduodenal administration of istradefylline at 30 and 100mg/kg. The applicant considers it unlikely that this is a result of the pharmacological action of istradefylline but adenosine A_{2A} receptors are present in skeletal muscle vasculature and when interstitial adenosine concentrations increase secondary to elevated metabolism (e.g., during exercise), the vascular A_{2A} receptors can promote vasodilation. Furthermore, according to Lynge and Hellsten (2000), A_{2A} and A_{2B} but not A_1 receptors are located in human skeletal muscle membrane and cytosol, while all the 3 receptor types were found in vascular smooth muscle cells and in microvascular endothelial cells: through the different actions (inhibitory and stimulatory) on A_2 and A_1 receptors, adenosine regulates muscle blood flow. The applicant considers the metabolic demand for increased skeletal muscle metabolism and blood flow would have been negligible under the conditions of this study and adenosine is one of several factors responsible for the coupling of skeletal muscle metabolism and blood flow, hence the applicant considers it is unlikely that A_{2A} receptor blockade would have had any influence on femoral blood flow. The applicant suggests that gradual cooling of the legs as the experimental preparation deteriorates could reduce an already low metabolic demand in the vascular bed supplied by the femoral artery in an anesthetized dog preparation, resulting in gradual reductions in femoral blood flow. Although this does not explain why the finding occurred in the 30 and 100mg/kg dose groups but not in the 10mg/kg or vehicle control groups. However, the findings of this study were not considered reliable since only single doses were tested, the dogs were under general anaesthesia, and considering the gradual deterioration of the experimental preparation. Even if adenosine is only one of several factors responsible for the relatively close coupling between skeletal muscle metabolism and blood flow (Marshall, 2007; Mortensen, 2014), a toxicity finding of myocyte vacuolation in in mouse carcinogenicity studies from 250 mg/kg is also reported. The myocyte vacuolation observed in carcinogenicity studies as a clear effect of long-term exposure to istradefylline, appears not associated with an inflammatory response or other types of lesions. Therefore, although the mechanism of the effects of istradefylline on rodent skeletal muscle remains unknown, it is acknowledged that from a clinical point of view, no safety signals concerning skeletal muscle were observed, therefore, it's plausible to believe that istradefylline presents a low safety risk to the skeletal muscle function in humans.

Istradefylline alone had no effect on gastric emptying. L/C alone decreased gastric emptying. Istradefylline, co-administered with L/C, also decreased gastric emptying, suggesting that the decreased gastric emptying observed after istradefylline dosing with L/C should be attributed to the effect of L/C.

In summary, safety pharmacology studies indicate a significant CNS response of increased spontaneous locomotor activity following oral administration of istradefylline in rodents and common marmosets. Additionally, shortened pentobarbital-Na-induced sleeping time occurred in mice and increased rectal temperatures occurred in mice, rats and rabbits. Cardiovascular safety pharmacology studies indicate increased heart rate and blood pressure in conscious rats following oral istradefylline administration but this effect was attributed to an increase in spontaneous locomotor activity and it was not replicated in conscious dog studies, although locomotor activity levels were not reported. A finding of reduced femoral blood flow in anaesthetised dogs is of uncertain relevance and may be a result of the experimental preparation rather than a treatment-related effect.

Pharmacodynamic drug interactions

No studies were provided by the applicant.

2.3.2. Pharmacokinetics

Absorption

The plasma concentrations and pharmacokinetics of istradefylline after single oral or intravenous administration were studied in mice, rats, dogs, rabbits, marmosets and monkeys. In male mice administered a single oral dose of 0.3, 3 or 30mg/kg istradefylline, exposure increased greater than dose-proportionally with increasing dose. Istradefylline appeared to be rapidly absorbed at lower doses with a t_{max} of 0.5, 1 at 0.3 and 3 mg/kg respectively but absorption slowed at the higher dose, with a t_{max} = 5 hours at 30mg/kg. Although, systemic exposure was not considered saturated over this dosing range in mice. In rats administered a single oral dose of 0.3, 1, 3, 10, 30, 100mg/kg istradefylline, both area under the plasma concentration versus time curve from 0 to infinity ($AUC_{0-\infty}$) and C_{max} increased dose-proportionally up to 30 mg/kg ($AUC_{0-\infty}$=119, 294, 1078, 3046, 9549 ng · h/ml respectively) and bioavailability was calculated at between 19.7 - 25.7% in this dose range. However, exposure at 100mg/kg ($AUC_{0-\infty}$=11,480 ng · h/ml) was similar to the 30mg/kg, indicating that absorption had plateaued and bioavailability was calculated at only 7% at 100mg/kg. t_{max} ranged between 1.3 and 2.8 hours and $t_{1/2}$ for all doses up to 10 mg/kg was in the range of 1.89 to 3.1 hours, which was considered equivalent to the $t_{1/2}$ following a single iv dose (0.3mg/kg iv, $t_{1/2}$ = 3.07h) but $t_{1/2}$ was longer at 30mg/kg and 100mg/kg (4.1 h & 5.83 h respectively). Plasma concentrations of istradefylline were also measured following single iv administration of istradefylline at 0.3mg/kg in rats, results indicated $AUC_{0-\infty}$, clearance (CL), and volume of distribution steady-state (Vdss) were 463 ng·h/mL, 0.681 L/h/kg, and 2.39 L/kg, respectively, with a hepatic extraction ratio of 20%.

In rabbits, plasma concentrations of istradefylline increased more slowly following oral administration of a 100mg/kg dose, a C_{max} of 4826.7 ng/mL is reported 4 hours post-dose. A 300mg/kg dose level was included in this study but 1 rabbit died with a very high plasma istradefylline concentration (C_{max} = 21,722 ng/ml versus 5944 ng/ml in the surviving animal) leaving only n=1 in the group, therefore the data from this group are not considered robust. The $t_{1/2}$ and $AUC_{0-\infty}$ at 100mg/kg were 6.79 hours and 46,282 ng·h/mL, indicating that the exposure of rabbits to istradefylline was higher than that seen in rats and dogs after oral administration of the same dose. In dogs, plasma concentrations of istradefylline were measured following single oral administration of 0.3, 3, 30, and 100 mg/kg of istradefylline. $AUC_{0-\infty}$ and C_{max} increased less than dose proportionally with increasing dose and oral bioavailability was calculated at 106.9, 52.9, 28.3 and 18.8% respectively. t_{max} was reported at 0.6, 1.5, 2.0 and 8.7 h respectively, but the average t_{max} at 100mg/kg was considered the result of an aberrant value in this group (n=3, individual t_{max} values = 1h, 1h & 24h). A second peak in the plasma-concentration time profile was observed in 2 out of 3 dogs following administration of 30 and 100 mg/kg, this second peak corresponded to the C_{max} for 1 dog in the 100mg/kg group. As a result of this second peak in the plasma concentration-time profile, the $t_{1/2}$ showed wide variation and the measured $t_{1/2}$ was not considered a valid estimate of $t_{1/2}$ for this study. Plasma concentrations were also measured in dogs after intravenous administration of 0.1 mg/kg istradefylline, the dose level was limited by the poor water solubility of istradefylline and therefore plasma concentrations of istradefylline could only be determined for 2 hours post-dose. The values for $t_{1/2}$, $AUC_{0-\infty}$, CL, and Vdss were 1.43 hours, 58 ng· h/mL, 1.84 L/h/kg, and 2.88 L/kg, respectively.

In female cynomolgus monkeys, following single oral doses of 10 and 30 mg/kg istradefylline, C_{max} values of 100.5 and 137.6 ng/mL were reached after 6.7 and 4.7 hours, respectively. At 30 mg/kg Mean $t_{1/2}$ and $AUC_{0-\infty}$ were 5.88 hours and 2468 ng· h/mL respectively but high variability and low AUC values compared to other species indicate exposure in cynomolgus monkeys is variable and istradefylline is poorly absorbed in this species. In male and female MPTP-treated common marmosets, following oral administration of 0.3, 1, 3, and 10 mg/kg istradefylline, C_{max} and AUC_{0-72} increased with increasing dose in a dose-proportional manner and t_{max} was prolonged as the dose increased.

The plasma concentrations and PK of istradefylline were also reported from 4-week repeat-dose toxicology studies in rats and dogs, the toxicokinetics following daily oral administration in these studies are included in separate study reports. Both male and female animals were included in both species, allowing for the assessment of gender-based differences in PK. The sponsor has also included studies using both micronized and un-micronized istradefylline in rats allowing for comparison, although different dose levels were used in the two studies. In both rat studies istradefylline was administered orally once a day for 4 weeks to male and female CD rats; micronized istradefylline was dosed at 25, 100, and 400 mg/kg/day (#KHK33/960498) and non-micronized istradefylline was dosed at 6, 30, 160, and 800 mg/kg/day (#A-95-133). For the toxicokinetic analysis, plasma concentrations of istradefylline were measured on Days 1 and 28 (#KHK33/963444 and #A-96-139). C_{max} and AUC_{0-24} increased with increasing dose over the dose range in both studies, although the increases were less than dose proportional, hence istradefylline did not accumulate during repeated dosing in male and female rats. No gender-related differences are reported, however, decreased exposures occurred in the AUC_{0-24} of female rats on day 28 at 100mg/kg (AUC_{male} = 51,800 ng· h/ml, AUC_{female} = 29,105 ng· h/ml) in the micronized istradefylline study, and at ≥30mg/kg in the non-micronized istradefylline study (30mg/kg: AUC_{male} =11,482 ng· h/ml, AUC_{female} = 6,896 ng·h/ml; 160mg/kg: AUC_{male} =50,146 ng·h/ml, AUC_{female} = 28,126 ng·h/ml; 800 mg/kg: AUC_{male} = 99,836 ng·h/ml, AUC_{female} = 65,436 ng·h/ml). In general, exposures to istradefylline in the micronized istradefylline study at doses of 100 mg/kg and 400 mg/kg were similar to that obtained in the non-micronized istradefylline study at doses of 160 mg/kg and 800 mg/kg respectively, indicating that micronizing istradefylline increased exposure as anticipated. The micronized istradefylline toxicokinetics study report (#KHK33/963444) is GLP-compliant but the non-micronized toxicokinetics study report (#A-96-139) does not include a GLP compliance statement, although the corresponding toxicology study report (#A-95-133) is GLP-compliant. The applicant clarified that the toxicokinetics reported in study #A-96-139 was not conducted in compliance with GLP but provided a justification that the final study reports were audited by the Kyowa Kirin Co., Ltd. Quality Assurance Unit at its GLP facility and considered to have accurately described the test method and results.

In dogs, istradefylline was administered orally once a day for 4 weeks either as micronized istradefylline at 30, 100, and 300 mg/kg/day (#KHK32/961196) or as istradefylline micronized with hydroxypropylmethylcellulose at 25, 100, and 400 mg/kg/day (#A-96-83). For the toxicokinetic assessments, plasma concentrations of istradefylline were measured on Days 1 and 28 (#KHK32/962036), or Days 1 and 23 (#A-96-29). The C_{max} and AUC_{0-24} increased with increasing dose over the studied dose range and the increase in C_{max} was less than dose-proportional but the increase in AUC_{0-24} was generally dose-proportional. No evidence of gender-related differences in exposure are reported but there is evidence of accumulation, with C_{max} and AUC_{0-24} higher on day 28/ 23 following repeated dosing when compared to single dosing on day 1. The toxicokinetics study report #KHK32/962036 (corresponding to toxicology study #KHK32/961196) is GLP-compliant but the toxicokinetics study report #A-96-29 does not include a GLP compliance statement, although the corresponding toxicology study report (#A-96-83) is GLP-compliant. The applicant clarified that the toxicokinetics reported in study #A-96-29 were not conducted in compliance with GLP but provided a justification that the final study reports were audited by the Kyowa Kirin Co., Ltd. Quality Assurance Unit at its GLP facility and considered to have accurately described the test method and results.

A study in rats using radio-labelled istradefylline (#96-093) demonstrated that istradefylline was absorbed throughout the entire gastrointestinal tract. A 3 mg/kg dose of 14C-istradefylline was administered to ligated loops of gastrointestinal tract and the remaining radioactivity after 3 hours was subtracted from the total radioactivity administered to each section to calculate the systemically absorbed fraction for each section; the duodenum (39.5%) and small intestine (37.4%, 40.2%, 35.5 % for upper, middle and lower respectively) were the major sites of absorption > rectum (25.6%) > colon (16.5%) > stomach (7.0%).

Distribution

Tissue distribution studies in rat were conducted using both a tissue dissection, homeogenization and solubilisation method (#96-217), and whole-body autoradiography (#96-308). In both studies, a single dose of 3 mg/kg 14C-istradefylline was administered orally and tissue distribution was assessed at 2, 6 and 48 hours post-administration. In general, the highest concentration of radioactivity in tissues and organs was recorded at 2 hours post-dose using both methods, and the radioactivity in most tissues and organs subsequently decreased in parallel with the decrease in plasma radioactivity in both studies.

For the tissue dissection study (#96-217), the sponsor reports the proportion of radioactivity as highest in perirenal fat (19.05% of dose) > muscle (7.29% of dose) >small intestine (6.90% of dose) > skin (6.34% of dose) > liver (4.58% of dose) > other tissues (including plasma and blood; <1% of dose). However, there is an issue with the calculation of the distribution of radioactivity (% of dose) in the study report for which a correction is included in Final Report Amendment No. 3, although the final study report text has not been updated and the revised table appears to still contain errors. The distribution of radioactivity 2 hours post-dose is more accurately reported as highest in the small intestine (T/P = 29.56) > perineal fat (T/P = 21.72) > brown fat (T/P = 12.89) > liver (T/P = 7.82) > adrenal gland (T/P = 5.53) > harderian gland (T/P = 5.22) > large intestine (T/P = 5.17) > stomach (T/P = 4.50) > kidney (T/P = 3.10) > pancreas (T/P = 3.03) > other. At 48 hours post-dose, the highest radioactivity was observed in the small and large intestines (T/P = 86.47, 106.22 respectively)> liver (T/P = 29.32) > thyroid gland (T/P = 12.17) > kidney (T/P = 4.36) > other. Of relevance to the potential for phototoxicity, skin radioactivity concentration (864 ng eq./ml) was slightly higher than plasma (526 ng eq./ml, T/P = 1.64) at 2 hours post-dose and approximately equivalent at subsequent timepoints (6 h: Skin = 344 ng eq./ml, Plasma = 311 ng eq./ml). This slight early tendency of 14C-istradefylline to distribute to the skin has not been further addressed and no pigmented rat study is included in the pharmacokinetics package although an assessment of phototoxicity was completed in Long-Evans pigmented rats as part of the toxicology package which is acceptable. Total concentration of radioactivity in the eye was less than plasma at all timepoints and was undetectable at 48 h post-dose.

From the autoradiography study (#96-308), the highest levels of radioactivity were seen in white fat (Tissue to cardiac blood relative concentration ratio, T/B = 5.57) and the adrenal gland (a target organ of toxicity, T/B = 5.43) > brown fat (T/B = 4.17), harderian gland (T/B = 4.16) > liver (T/B = 4.13) > renal cortex (T/B = 3.10) > pancreas (T/B = 2.19) > testis (T/B = 1.68) > submaxillary gland (T/B = 1.57) > heart (T/B = 1.46) > spleen & lung (T/B = 1.28) > renal medulla (T/B = 1.12) > pituitary (T/B = 1.05) . The lowest levels of radioactivity reported were observed in the thymus (T/B = 0.59), brain (T/B = 0.65), muscle (T/B = 0.76) and bone marrow (T/B = 0.86), with none detected in the eyeball. At 48-hours post-dose radioactivity was below the limits of detection in the tissues and organs with the exception of the gastrointestinal tract which still had measurable radioactivity as a result of enterohepatic recirculation, as supported by a study in bile duct–cannulated rats included in the excretion section (#2003-093A).

Protein binding of 14C-istradefylline was assessed *in vitro* in rat, pregnant rabbit and dog plasma, 3H-istradefylline binding was assessed in human plasma. Protein binding was generally unchanged over the concentration range of 10 – 10,000ng/mL tested in each species, ranging from 95.6 to 96.1% in rat serum (d-04-494), from 97.8 to 98.1% in pregnant rabbit serum (d-06-090), from 90.9 to 92.5% in dog serum and from 95.0 to 96.6% in human serum (#99-159A), indicating a similar unbound fraction in humans and rats, which is twice the unbound fraction in rabbits and half the unbound fraction in dogs. *In vivo*, protein binding was assessed in rat serum and dog's plasma following oral administration of 3 mg/kg of 14C-istradefylline at 0.5, 2 and 6/8 h post-dose. In agreement with the *in vitro* findings, in rats the protein binding of radioactivity was 95.1%, 94.1%, and 92.1%, at 0.5, 2, and 6 hours post-dose, respectively. In dogs, the protein binding of radioactivity was 94.0%, 91.8% and 90.7% at 0.5, 2

and 8 hours post-dose, respectively. Decreases in protein binding over time are attributed to the presence of metabolites which is plausible.

Tissue distribution and Foetal Transfer studies in pregnant rats were conducted using both a tissue dissection method (#96-644A), and whole-body autoradiography (10591 #96-643A). In both studies, a single dose of 3 mg/kg 14C-istradefylline was administered orally on gestational day (GD) 12 or 19 and tissue distribution was assessed at 2, 8 and 24 hours post-administration. In general, on both GD 12 and day 19 the radioactivity in all maternal tissues was the highest at 2 hours post-dose and decreased thereafter.

For the tissue dissection method (#96-644A), following administration on GD12, the ratios of the radioactivity in the whole foetus to the maternal intracardiac blood were 0.7, 0.5, and 0.6 at 2, 8, and 24 hours respectively. However, following administration at GD19, the ratios of radioactivity in the whole foetus to the maternal intracardiac blood were 1.2, 1.7, and 3.3 at 2, 8, and 24 hours respectively. Consistent with these data, results from the whole-body autoradiography method (10591 #96-643A), demonstrate following administration on GD12, radioactivity in the foetus was 0.1-fold and 0.3-fold that of maternal intracardiac blood at 2 and 8 h post-dose respectively, while radioactivity was below the limit of detection 24 hours after administration. While similarly to the previous study, on GD19 the radioactivity in the foetus was 1.2-fold, 1.6-fold, and 3.5-fold higher than that in maternal intracardiac blood, respectively. The radioactivity in the foetus was eliminated more slowly than that in maternal intracardiac blood.

These data indicate significant foetal exposure to istradefylline and/ or metabolites which is higher on GD19 than GD12, indicating that placental transfer of istradefylline occurred more easily at the later stage of pregnancy than during organogenesis.

Metabolism

The plasma metabolite profile of istradefylline was investigated by radiochromatography in male CD-1 (ICR) mice (d-04-409), male CD (SD) (d-04-200) and Hannover Wistar (BrlHan:WIST) rats (d-04-362), and male beagle dogs (d-04-401). 3 mg/kg 14C-istradefylline suspended in 0.5 w/v% methylcellulose was administered orally in each species and assessments were made at 1, 2, and 6 hours post-dose in mice, 2 and 6 hours post-dose in rats, and 0.5, 2, and 8 hours post-dose in dogs. The major components in the plasma of all three species was istradefylline (45 to 55% in mice, 40 to 57% in rats, 44 to 75% in dogs). The next major peak in both mice and rats was M5, the glucuronide conjugate of 4'-O-demethyl istradefylline (36 to 42% in mice, 17 to 25% in rats). In mice, the minor metabolites M1, M4, and M8 were also detected at < 5% of the total radioactivity. M1 plasma content was slightly higher in rats at approximately 11% of total radioactivity, but other minor metabolites in rats similarly included M4 and M8 detected at < 3% of the total radioactivity. The most abundant components after istradefylline in dog plasma were M4 and M5, which comprised 10% and 2%, 18% and 4%, and 19% and 13% of the total at 0.5, 2, and 8 hours post-dose, respectively. In addition, M1, M8, the cis isomer of istradefylline, the glucuronide conjugate of M1* and the glucuronide conjugate of M9 (4'-O-demethyl-hydrogenated istradefylline) (M1 1= 4'-O-demethyl-hydrogenated istradefylline glucuronide) were detected as minor metabolites in dogs. Hence, in mice, rats and dogs, 4'-O-demethylation followed by glucuronide conjugation was considered to be the main metabolic pathway.

The metabolite profile of istradefylline in urine and faeces, was also investigated by radiochromatography following similar 3mg/kg oral 14C-istradefylline dosing in rats (d-04-200) and dogs (d-04-401). By 48hrs post-dose in rats, 18% of the total radioactivity in the administered dose had been excreted into urine and 68% into faeces. Major metabolites in the urine were M3 and M1 at 5% and 2% of the total dose respectively, unchanged istradefylline was not detected in urine, indicating that renal clearance of istradefylline did not occur in rats. Major metabolites in the faeces were M3 (31%), M1 (9%) and M10 (3', 4'-O-didemethyl-hydrogenated istradefylline) (8%), with unchanged istradefylline detected at just

1.62% of the radioactivity of the total administered dose. These data, together with an oral bioavailability in rats of 23.3%, indicate that istradefylline undergoes first pass metabolism in rat stomach/ intestines. In dogs the percentage of the total radioactivity in the administered dose excreted by 48 hours in urine was 5%, and in faeces was 83%. Major metabolites in the urine were M4, M5, M11, and M1, which represented 1.4%, 0.6%, 0.4%, and 0.4% of total dose respectively, unchanged istradefylline was detected at only 0.08%. Major components in faeces were istradefylline, M10, and M1, these comprising 28.3%, 26.4%, and 11.9%, respectively, of the total radioactivity in the dose. Oral bioavailability in dogs was 52.9%. In addition, metabolites in bile were also studied in rats (#2000-372A). Biliary excretion of radioactivity accounted for 41%, 87%, and 94% of the total radioactivity in the administered dose, at 6, 24, and 48 hours post-dose respectively, with M5, M4, M1, and an unidentified metabolite detected in bile excreted within 24 hours at 40.5%, 31.4%, 4.0%, and 8.7% of dose respectively. The metabolite M3, which was the main metabolite in rat urine and faeces, was not detected in bile. Since M1 was metabolized to M3 in the intestinal content homogenates (study #98-072A), the radioactivity excreted as M5 and M4 in the bile was most likely hydrolysed to M1 and further metabolized to M3 in intestinal contents.

The metabolite profile of istradefylline in the brain, was also investigated by radiochromatography following similar 3mg/kg oral 14C-istradefylline dosing in rats (#2000-140A) at 0.5, 2 and 6-hours post-dose. Istradefylline accounted for 86% to 80% of the total radioactivity in the administered and M1 accounted for 8 to 16% at 0.5 to 6 hours after administration. M3 accounted for 3.2% and 0.7% at 0.5 and 6 hours post-dose, respectively, but was not detected at 2 hours post-dose. The brain-to-plasma concentration ratios (Kp) for istradefylline and its metabolites ranged from 1.69 to 2.21 for istradefylline and from 1.42 to 2.85 for M1 at 0.5 to 6 hours after administration, with permeability of the brain to M5 and M4 extremely low (Kp for M5 = 0.00185 to 0.0213, no M4 was detected in the brain).

Summary data from six human volunteers administered 40mg istradefylline (XBL03093, #RPT01051) indicates that istradefylline was the major component in the plasma, detected 2 hours post-dose at 75.6% of total radioactivity, reducing to approximately 50% up to 36 hours post-dose. The next highest peaks were M5 (detected at 3.4 to 24.8% of total), M8 (3.8 and 12.6% of total), M11/M12 (M12 3', 4'-O-didemethyl-hydrogenated istradefylline monoglucuronide) (ND/2.6 to 24% of total), M4 (ND/6.4 to 14.2% of total), M1 (ND/3.6 to 6.8% of total). Of the total radioactivity in the administered dose 17 to 40% was excreted into urine and 19 to 56% into faeces. Unchanged istradefylline was not detected in human urine but was detected at 1.25 to 31.55% of radioactivity in administered dose in human faeces. These data indicate no qualitative species differences in the metabolism of istradefylline.

The potential of istradefylline to induce hepatic drug metabolizing enzymes was investigated in rat liver microsomes from Male CD (SD) rats administered 1, 30, and 800 mg/kg/day of istradefylline suspended in 0.5 w/v% methylcellulose orally for 7 (7L859) with phenobarbital (80 mg/kg/day) administered as a positive control. Cytochrome P450 contents and the activities of aminopyrine N-demethylase and 7-ethoxycoumarin O-deethylase decreased significantly in the high dose group, but it was considered unclear whether this was a treatment-related effect or an indirect result of the toxicity at this dose level (2 rats in the high dose group died). Despite these inconclusive findings, there were no significant increase in enzyme activities reported, indicating that istradefylline did not induce hepatic microsomal drug-metabolizing enzymes.

A second study examining the effect of istradefylline on UGT in rat liver (P050954) was conducted to assess a possible correlation between treatment-related effects on circulating thyroid hormone levels and UGT activity. Male and female Hannover Wistar (BrlHan:WIST) rats administered 100 and 320 mg/kg/day of istradefylline suspended in 0.5 w/v% methylcellulose orally for 4 consecutive weeks, with phenobarbital (80 mg/kg/day) administered as a positive control. Plasma concentrations of thyroid hormones (TSH, total T4, and total T3) and UGT activity were measured and no significant findings are

reported with the exception of a small decrease in T4 in males from the 320-mg/kg group which was not considered biologically significant.

However, in sandwich-cultured human hepatocytes, istradefylline at concentrations from 0.1 to 30 µmol/L did not induce CYP1A2 and CYP2B6 messenger RNA (mRNA) levels, but caused concentration-dependent increases in CYP3A4 mRNA levels (XT-153111), suggesting that istradefylline is likely an *in vitro* inducer for CYP3A4 which is of clinical relevance for drug-drug interactions.

Studies conducted with human metabolic enzymes, liver microsomes, and hepatocytes are included in the clinical assessment section. In summary, these data indicate istradefylline was primarily metabolized by CYP1A1, CYP3A4 and CYP3A5 to demethylated metabolites and by CYP3A4 and CYP3A5 to hydroxylated metabolites. In addition, istradefylline inhibited CYP3A activities in a mechanism-based, concentration dependent and time-dependent manner. No effect on other CYP isozymes are reported.

Excretion

The excretion of radioactivity after the oral administration of 3 mg/kg 14C-istradefylline suspended in 0.5 w/v% methylcellulose was investigated in male CD (SD) rats (#96-158) and male beagle dogs (#96-218). In rats, of the total radioactivity in the administered dose, the percentages recovered in the urine and faeces up to 168 hours post-dose were 24.9% and 70.3%, respectively. In dogs, radioactivity was eliminated rapidly in urine and faeces, with 6.4% and 85.9% recovered up to 48 hours post-dose respectively, increasing to 6.8% and 92.7% of the total radioactivity respectively, at 168 hours. The total recovery of the radioactivity was 96.4% in rats and 100.0% in dogs and the main route of excretion of the radioactivity was the faeces in both rats and dogs. Summary data from clinical study (XBL03093, #RPT01051) were also presented for comparison with 38.9% recovered in urine and 48.0% recovered in faeces, indicating that the excretion pattern is somewhat different in humans with excretion via the faeces less dominant, but total recovery of radioactivity was also lower in humans at 86.9%. Furthermore, in a similar study conducted in male bile duct-cannulated CD (SD) rats (#96-194), cumulative excretions of radioactivity in bile, urine, and faeces up to 48 hours post-dose were 77.5%, 6.9%, and 12.6% respectively, and the total recovery of radioactivity was 100.6 ± 7.3% including the radioactivity in the gastrointestinal contents and the carcass. Indicating that the main route of excretion of radioactivity was in the bile of these rats. In a study designed to examine enterohepatic recirculation, bile duct cannulated rats were administered 3 mg/kg of 14C-istradefylline suspended in 0.5 w/v% methylcellulose and bile was collected 24 hr post-dose. This bile was then administered as a single intraduodenal dose to other bile-duct cannulated rats (#2003-093A) and biliary excretion of radioactivity was 75.8% at 48 hours post-dose, while urine and faecal excretion were 7.3% and 17.3% respectively. At 48 hours post-administration, the percentage of the radioactivity remaining in the gastrointestinal contents was 0.3% and the total recovery of radioactivity was 100.7%. The reabsorption ratio, calculated from the sum of the cumulative excretion of the radioactivity in urine and bile and the remaining amount of the radioactivity in the carcass, was 83.1% of the dose.

In summary, following the oral administration of 3 mg/kg of istradefylline, approximately 20% of the dose was absorbed as the intact drug (rat oral bioavailability at 3mg/kg = 23.3%). The remainder of the administered dose likely underwent pre-systemic metabolism (1st pass effect) and was metabolized or degraded in the stomach and/or by microorganisms present in intestinal contents. Approximately half of the radioactivity, following oral administration, was reabsorbed by enterohepatic recirculation with the remainder excreted in the faeces.

Of note, the transfer of radioactivity into milk was investigated in lactating rats following oral administration of 3 mg/kg 14C-istradefylline 9 days post-delivery (#98-654A). Maternal C_{max} of 419 ng eq/mL occurred at 2.8 h post dose, and t1/2 was 16.1 h, with an $AUC_{0-\infty}$ of 5220ng eq·h/mL. In the milk, a C_{max} of 4590 ng eq/mL occurred 1 h post-dose and decreased thereafter and t1/2 was 4.6 h, with an $AUC_{0-\infty}$ of 30,300 ng eq·h/mL, which is 5.8-fold higher that the radioactivity in maternal blood. Hence

the apparent milk to blood partition coefficients were 2.6 to 12.7, at 0.5 to 24 h post-administration. Furthermore, radioactivity in the blood of suckling rats increased with time, with a C_{max} of 79 ng eq/mL at 12 hours post-administration to the maternal lactating rats, which could still be detected up to 48 hours afterwards. The blood concentration of radioactivity in suckling rats was 0.7-fold that of lactating rats, and 0.02-fold the C_{max} of radioactivity in milk from lactating rats 12 hours post-administration. The radioactivity was eliminated from the blood of suckling rats more slowly than from that of the maternal lactating rats.

Pharmacokinetic drug interactions

In order to assess the effect of co-administration of L/C on the absorption of istradefylline, a single dose iv administration study (#2001-401A) and a single dose oral administration study (#2001-567A) were conducted in rats. Plasma concentrations of istradefylline and M1 were measured following intravenous administration of 0.3 mg/kg of istradefylline (solution in 40 v/v% dimethylacetamide) to vehicle- or L/C (250/25 mg/kg)-treated male CD (SD) rats and plasma concentration-time profiles were similar between istradefylline alone and the combined administration groups. No significant differences in PK parameters are reported indicating that L/C co-administration did not affect metabolism or distribution of istradefylline. However, when istradefylline (suspension in 1 w/v% methylcellulose) was orally administered in combination with L/C, the AUC_{0-t} values for istradefylline increased to 105, 119, and 134% compared with that in the group that received istradefylline alone at 1, 3, and 100 mg/kg, respectively. These data indicate that L/C co-administration affected the absorption or istradefylline. It is considered to be the result of a dopamine-mediated reduction in intestinal motility leading to prolonged intestinal residence time of istradefylline. This affect is in agreement with findings reported in the toxicology section, indicating increased systemic exposure to istradefylline when L/C is co-administered, although this may be a species specific effect in rats as it was not replicated in the clinical data when comparing PD patients receiving istradefylline and concomitant L/C versus healthy volunteers receiving istradefylline alone.

Of note, drug transporter studies conducted using Caco-2 and human transporter expressing cells indicate istradefylline is an inhibitor of human P-glycoprotein (P-gp) and BCRP (breast cancer resistance protein) and the drug-drug interaction (DDI) risk assessment indicates that this effect may also occur *in vivo*. This issue will be addressed in the clinical pharmacology section. Further *in vitro* studies indicate an inhibitory effect of istradefylline on OATP1B1, OATP1B3, OAT1, OCT2, MATE1, and MATE2-K but this is not predicted to be relevant *in vivo*.

Other Pharmacokinetic studies

When comparing data from the two research facilities where the istradefylline dogs studies were conducted, HLS contract research organization and Kyowa Hakko Kogyo's Toxicological Research Laboratories, an effect noted regarding variability in systemic exposure. There was also a difference in feeding schedules between these two sites. To assess the effect of differences in feeding time on systemic exposure, an oral dose of 100 mg/kg of istradefylline was administered at 9:00 a.m., with food subsequently given according to two different feeding schedules (#A-03-089). The mean C_{max} and AUC_{0-t} values in animals fed from 10 a.m. to 5 p.m. tended to be higher than the means in animals fed from 3 p.m. to 9 a.m. These results indicate that feeding time affected the absorption of istradefylline in dogs.

2.3.1. Aetiology and pathogenesis

The etiology of PD is unknown. Most motor symptoms of PD result from the progressive degeneration of the dopaminergic neurons in the substantia nigra pars compacta (SNc). As degeneration occurs, striatal concentrations of dopamine decrease, leading to reduced stimulation of dopamine receptors in the striatum. Dopamine activates the 'direct pathway' from the striatum of the basal ganglia via D1 receptors

and suppresses the 'indirect pathway' from the striatum via D2 receptors. Thus, the reduced striatal dopamine that occurs in PD results in decreased activity of the direct pathway and increased activity of the indirect pathway. Increased excitability of the indirect pathway can be mediated by adenosine A_{2A} receptors in the striatum. The imbalance of activity between the direct and indirect pathways in PD is thought to result in the hallmark symptoms of bradykinesia, rigidity, tremor, and loss of postural reflexes.

2.3.2. Toxicology

The applicant has submitted an extensive safety package for Istradefylline in line with the requirements of the respective ICH guidances. A summary of the presented data is presented below:

Single dose toxicity

GLP compliant single dose toxicity studies were conducted in mouse, rat, dog and monkey. These studies report Istradefylline to exhibit low toxicity following acute administration with no mortalities reported in any species. The primary findings consisted of increase activity and piloerection in rodents, with decreased respiration and lethargy were reported in the mouse study with no histopathological correlates. In dog and monkey no toxicity was noted with grey/white faeces the primary finding likely resulting from excretion of unabsorbed test-article. PK data acquired in dogs shows exposures rise in a less than dose-proportional manner with no significant sex related differences in exposure.

Repeat dose toxicity

Repeat dose toxicity for Istradefylline was assessed in Rat and Dog in studies of up to 26 and 52 weeks duration respectively. In addition, studies assessing toxicity when Istradefylline is co-administered with C/L were also assessed in studies of up to 13-weeks duration.

Rat Studies

Overall, 7 pivotal GLP repeat-dose toxicity studies were conducted in rats (3 studies, 4 to 26 weeks) and beagle dogs (4 studies, 4 to 52 weeks) using two istradefylline formulations (non-micronized or micronized with or without hydroxypropylmethylcellulose). In addition, a number of GLP-compliant studies to assess the toxicity of istradefylline when administered in combination with levodopa and carbidopa as per the proposed indication have also been submitted. It should be noted that in the 26-week repeat-dose toxicity study in rats, plasma samples were only acquired 4 and 24 hours post dose on day 1, week 13 and week 26, although these data confirm exposure to the test-article, no C_{max} or AUC data are available from this study. AUC data from the 4-week study also utilising the non-micronised drug substance are therefore used for exposure margin comparisons. The applicant has presented a side by side comparison of plasma exposures at 4 and 24 hours post dose from 4 and 26-week studies. These data suggest higher exposures in the 26-week study, particularly 24 hours post dosing suggesting some degree of accumulation over dosing. The exposure margin comparisons provided by the applicant are not considered accurate on this basis.

Clinical signs in these studies included increased locomotor activity which resolved to some degree in all studies following repeat dosing. Of note, this finding did not appear to be dose-related. Minor increases in food consumption and body weight/body weight gain were noted in the 26-week study.

The primary target organs identified in these studies were the adrenals, kidneys, liver, pancreas and lung.

The adrenal changes were characterised by hypertrophy of adrenal zona fasciculata cells in high dose groups and were not accompanied by any progression of lesions to neoplasia or adrenocortical degeneration/necrosis up to the highest doses administered to rats (320 mg/kg/day) in the 2-year

carcinogenicity study. Additional mechanistic studies were undertaken to assess the mechanism for these effects (see under 'Effects on adrenal related hormones' below).

Pancreatic cell vacuolation and apoptosis/single cell necrosis were reported in several rodent studies as well as the mouse 2-year carcinogenicity study. This was considered linked to feeding status. No mechanistic studies examining the cause of these findings were performed. However, in the absence of a clear clinical signal it is accepted that these findings are unlikely clinically relevant.

Increased liver weight was noted in several rodent repeat-dose toxicity studies with corresponding increases in total bilirubin reported in the 26-week study and in the kidneys, vacuolation of cortical tubule epithelial cells was reported.

Foci of vascular mineralisation were noted in the brains of animals in the high dose group in the 26-week repeat-dose toxicity study in line with findings in the rat carcinogenicity study. The applicant has presented a separate discussion on these findings, see below under carcinogenicity for a discussion on these findings.

Combination toxicity studies revealed that co-administration of Istradefylline with L/C is associated with a significant increase in Istradefylline exposures in rat. This resulted in premature treatment related deaths in animals administered the high dose combination (400/250/25 istradefylline/L/C mg/kg/day). Most of these deaths occurred in the first week of treatment and clinical signs in these animals included convulsions and rapid respiration. They were also associated with increases in alveolar macrophages and perivascular inflammation as well as acinar cell apoptosis in the pancreas and renal and thymus toxicity. Additional studies demonstrated that these findings were likely related to the increased exposures to istradefylline rather than a direct PD interaction. This is accepted. Of note, there were no histological changes in the brain noted in these studies, this is likely related to the short duration of these studies.

Dog Studies

The toxicity of Istradefylline both alone and in combination with levodopa and carbidopa has also been assessed in dog. Istradefylline was relatively well tolerated in dog with no test-article related mortality reported in any study. Initial 4 and 26-week studies in dog utilised a micronized 1:2 mixture of Istradefylline and HPMC. In line with rat studies, liver (hepatocyte hypertrophy), lung (increase in alveolar macrophages) and adrenals (cortical hyperplasia) were identified as target organs. Similar results were obtained in subsequent 4 and 52-week studies utilising micronized Istradefylline with adrenals and lung-identified as target organs. Effects on the lung (increased alveolar macrophages) were not present in recovery animals. Increase alanine aminotransferase (ALT), aspartate aminotransferase (AST), cholesterol and bilirubin were evident in most studies indicating potential liver toxicity. However, except for bile plugs in bile canaliculi observed in the 26-week but not the 52-week study these findings were without histopathological correlates.

Genotoxicity

Genotoxicity has been assessed in line with option 1 of the standard battery of genotoxicity testing outlined in ICH S2(R1). In addition, a test assessing Istradefylline induced unscheduled DNA Synthesis in Rat Liver has been conducted. All tests were conducted appropriately and reported negative results for mutagenicity in a bacterial reverse mutation assay and chromosomal aberrations in an *in vitro* mammalian cell ass and *in vivo* micronucleus assay in mice. It is accepted that the data presented are sufficient to conclude that Istradefylline is not genotoxic.

Carcinogenicity

The potential carcinogenicity of Istradefylline was assessed in two year studies conducted in rat and mice. Doses were based on dose range finding (DRF) studies of 13-weeks duration.

Findings in the mouse DRF study included increases in ALT and AST as well as histopathological effects on the liver, lungs and pancreas in line with the results of repeat dose toxicity studies conducted in dogs and rats. Skeletal muscle myocyte vacuolation and thymus atrophy were also noted which were not observed in studies conducted in other species. 250 mg/kg was identified as the maximum tolerated dose in this study and as the top dose in the subsequent pivotal carcinogenicity study in mouse. Only basic TK data from the mouse carcinogenicity study is available with samples acquired at 4 and 12 hours post dosing on weeks 13, 26, 52, 78 and 104. The analysis of these samples does reveal adequate exposures. At 4 hours post dose exposures increased in a less than dose proportional manner with a slight increase in exposure following repeated dosing evident. There was higher mortality in the 25 mg/kg/day male group relative to control, this occurred primarily in the initial 6 months of the study and was attributed to increased fighting between animals and the trend stopped following single housing. Increase mortality in males in the high dose groups was evident in the second year of the study, this did not reach statistical significance and it is not clear if this was a treatment related effect. Treatment had no other effect on the rate of mortality in this study. Non-neoplastic findings were similar to those observed in the DRF study and in line with those in repeat-dose toxicity studies in other species (with the exception of skeletal muscle vacuolation) with adrenals, pancreas, lung and liver identified as target organs. Foci of mineralisation in the brain primarily restricted to the thalamus were observed in all groups including controls. This was not associated with any neuronal degeneration, gliosis or inflammatory response. The incidence of this finding was slightly higher than historical control data but the findings were not dose related and it was concluded that in mouse this finding is related to normal aging and rather than the test-article. This is endorsed.

The number of mice with blood vessel tumours was higher in the female high dose group, as these are considered 'common' tumours in mice this did not meet the pre-specified level of statistical significance. This is accepted.

In the rat 13-week DRF study 2 animals died prematurely in the high dose group, one male was test article related and one female had a cause of death unknown. Findings in the DRF study included effects on the kidney, lung, adrenals, thymus and spleen. In the pivotal 104-weeek carcinogenicity study in rats TK data obtained 12 hours post dose on day 1, and week 13, 26, 52, 78 and 104 demonstrated adequate exposures throughout the duration of the treatment period. There was an increase in mortality which was statistically significant in both male (28/52) and female (35/52) high dose groups and considered treatment related. Lung lesions occurred in higher incidence in these groups and were considered related to the deaths of several animals with renal lesions also noted in some. Mortality in lower dose groups was not affected by treatment. A dose related increased in the incidence of hyperactivity and convulsions was observed in animals of both sexes in the mid and high dose groups. Non-neoplastic findings were in line with previous studies with target organs identified as adrenals, bone marrow (femur and sternum), heart (myocardial fibrosis, dose related severity), kidneys (nephropathy in mid and high dose females), liver (hepatocytic hypertrophy), lungs (inflammatory lesions), pancreas (acinar cell vacuolation/apoptosis), overaries (absent cornea lutea), skeletal muscle (myofibre degeneration), thymus (thymic cysts). In addition to these findings, a dose dependent and treatment duration related increase in the incidence and severity of foci of mineralisation in the brain vasculature was evident.

In the high dose group this finding was evident from week 24 and in the low dose group from week 39 (in decedent animals). This primarily consisted of single or scattered small foci (mostly minimal, i.e. 1-9 foci) of mineralization were observed mainly in the walls of small blood vessels in the basal ganglia, thalamus and mid-brain regions. This finding has been extensively characterised including the re-analysis

of brain sections form all other repeat-dose toxicity studies conducted. These analyses reveal this finding to be evident in the 13 and 26-week rat study. The applicant has provided a separate justification/risk assessment which argues that these findings are unlikely to be of clinical relevance. This is based on the species specificity of the findings with no similar findings noted in dog and no treatment-related findings noted in mouse (where this finding appears to be a natural age-related effect), and the lack of any corresponding histopathological evidence of gliosis (GFAP staining), inflammation or neurodegeneration/necrosis. Furthermore, there is no evidence from the existing clinical development or post-marketing safety database to indicate a significant risk of CNS related adverse events.

The applicant acknowledges that the detection of such adverse events (AEs) in the PD population is difficult and that no imaging data is available to confirm the that brain mineralisation does not occur clinically. Given the non-clinical data suggest this finding is dose and duration of treatment related and that it is not clear what effects such mineralisation would have clinically, it is considered unlikely that the existing clinical safety database would capture such a signal. It is accepted that the applicant has adequately investigated the finding and although no mechanism has been identified it is not clear that further studies would provide further reassurance. Brain mineralisation is considered a potential risk.

Analysis of tumours by trend test revealed an increase in mammary adenocarcinoma, thyroid gland adenoma and thyroid gland adenoma and adenocarcinoma which met the criteria for statistical significance for common tumours. The applicant further investigated whether the liver findings (increase in liver weight associated with liver enzymes increase with no histopathological correlates; centrilobular hyperthrophy of hepatocytes associated to liver enzymes increase; enhanced fatty changes of centrilobular zone associated with no abnormal changes in liver enzymes) observed in rat repeated-dose toxicity studies could promote development of thyroid tumours via increased metabolism of thyroxine and/or triiodothyronine which results in higher levels of thyroid stimulating hormones. Results from a 4 week study using istradefylline oral administration of 100 and 320 mg/kg/day no dose-related trends in the serum levels of total T3 or TSH was observed and the statistically lower T4 level in males in the 320-mg/kg group vs vehicle controls was within the range of vehicle controls thus considered to be not biologically relevant. Although results from this study are acknowledged, it is to be noted that 4-week duration maybe an insufficient time to trigger any alteration of the thyroid hormonal balance. However, considering that no thyroid related adverse events were noted in clinical trials, the potential safety risk in humans is expected to be low.

A statistically significant higher incidence of pancreatic islet cell adenoma was observed in the low dose male group, as a similar effect was not observed in other dose groups this was not considered treatment related, this is endorsed.

Reproduction Toxicity

Two GLP-compliant fertility and early embryonic development studies were conducted in rat. The first study examined the effect of Istradefylline on fertility and viability following oral administration of doses of 1, 6 and 30 mg/kg to male rats for a period of 9-4 weeks prior to mating and 4-weeks post mating and female rats 2 weeks prior to mating continued through to one-week post mating. There were no effects of Istradefylline administration noted in this study and therefore a second study utilising higher doses (to provide adequate exposure margins to clinical exposure) was conducted utilising doses of 160, 360 and 800 mg/kg. 160 mg/kg was set as the NOAEL based on decreased implantations at 360 mg/kg and decrease sperm motility and fertility index in the high dose group. No effect of treatment on embryonic viability was reported in these studies. No TK data were acquired in these studies. Although exposure data are available from a four-week toxicity study in rats (a-95-133-94-210).

The teratogenic potential of Istradefylline was investigated in embryo-foetal development (EFD) studies conducted in rat and rabbit. Doses in both studies were based on preliminary EFD studies undertaken in

pregnant animals with no teratogenic effects observed up to doses of 1000 mg/kg. In rat, doses of 0, 40, 200 and 1000 mg/kg non-micronised Istradefylline were chosen as the doses in the pivotal study. Clinical observations in this study report increased activity levels in all treated animals. A significant decrease in foetal body weight was noted in the high dose group. There was a significant increase in the incidence of skeletal variations in foetuses from the high dose group with spitting of the ossification centres of the central vertebral arch the most common finding with other vertebral skeletal variations noted in the high dose group. There was an increase in thymic remnants in the neck of foetuses in the treated groups but this was not statistically significant in pairwise comparison and was within historical control ranges. Although no significant differences in the incidence of visceral abnormalities was noted in this study, due to the high incidence of skeletal variations in the high dose group the findings at that dose level were considered equivocal with regard to teratogenicity and based on effects on foetal body weights was considered foetotoxic at the high dose level. Therefore, the NOAEL for developmental effects was set at 200 mg/kg in this study. No TK data have been provided for this study and no TK data from pregnant rats is available (see pre and post-natal development). The applicant has therefore used the PK data from female non-pregnant rats in the 4-week repeat dose toxicity study at 160 mg/kg/day to calculate/estimate exposure margins at the NOAEL of 200 mg/kg/day derived from the pivotal EFD study. The exposure margins calculated are not considered accurate.

In the preliminary rabbit study, doses of 1000 mg/kg were not tolerated in non-pregnant animals. Therefore, doses of 50, 200 and 800 mg/kg were used in the pivotal study. In the pivotal study, decreased body weight gain and food consumption were noted in dams in the high dose group. There was a decrease in foetal body weight, viability and placental weight in the high dose group relative to control. There was a non-statistically significant increase in skeletal variations in all treated groups in the main study with a significant increase in the incidence of delayed ossification of the phalanx of the forelimb in the high dose group. An increase in the incidence of external malformations was observed in all treated groups which was statistically significant in the high dose group. Istradefylline is considered teratogenic at high dose level with microphtalmia and oligodactyly observed in 2 and 3 foetuses respectively. It is noted that this occurred at dose levels at which maternal toxicity as evidenced through reduced food consumption and body weight gain was observed. Of note there was a non-statistically significant trend for increased incidence of visceral malformations in all treated groups. The reported NOAEL for effects on the foetus in this study was set at 200 mg/kg (which provides a ≈2.5-fold margin to exposures predicted at the higher dose proposed clinically). It should be noted that there was a numerically higher incidence of visceral malformations relative to control all dose levels in this study. However, this was not statistically significant and was almost within the range of fluctuation of background data, and therefore this was considered acceptable

These data indicate that Istradefylline can be considered foetotoxic and a potential human teratogen.

The effects of Istradefylline on pre and post-natal development were assessed in pregnant SD rats administered doses of 6, 25, 100 and 400 mg/kg micronized Istradefylline from day 7 of gestation to day 21 post-partum. No abnormalities or differences in the number of pups delivered, number of live pups, birth index or viability index were noted between control and treatment groups on Day 0. There were total litter losses in 2 dams in the control and 25 mg/kg group and 4 dams in the 100 and 400 mg/kg group during the lactation period. Lower mean viability was evident on days 0-4 in groups dosed in excess of 25 mg/kg. Viability was lower between days 4-22 in the high dose group with a lower number of live pups on day 4 in the 100 and 400 mg/kg groups. These differences were not statistically significant. There were no effects of maternal dosing on the F1 pups in any behavioural/functional endpoint assessed. There were no effects of maternal dosing on F1 reproductive performance and no abnormalities were observed in any F2 pups.

TK sampling in pre-natal and post-natal development toxicity study in pregnant rats was not measured. Exposure levels at 6 mg/kg (the NOAEL for F1) can be derived by using the AUC from non-pregnant

female rats in a 4-week+4 week of recovery repeated-dose toxicity study, for which at 6 mg/kg the AUC was 1922 ng*h/ml, even lower than clinical AUC at 40 mg/day (9817 ng*h/ml). Thus, no exposure margins exist for development effect in offsprings. Istradefylline causes developmental toxicity in rat.

Placental transfer of istradefylline occurred in pregnant rats with greater distribution to the foetus observed later in pregnancy. The ratio of foetal to maternal istradefylline exposure during the later stages of pregnancy was between 1.2 and to 3.5.

Istradefylline concentrations in milk were dose related and were 6.5 to 10-fold higher than in mother plasma. This is in line with results from PK study on excretion into milk in rat treated orally with istradefylline 3 mg/kg 9 days after delivery, in which the apparent milk to blood partition coefficients were 2.6 to 12.7, 0.5 to 24 hours after administration and $AUC_{0-\infty}$ of radioactivity in milk was 5.8-fold higher than that of the radioactivity in maternal blood. The blood concentration of radioactivity in suckling rats was 0.7-fold that of lactating rats. Radioactivity was eliminated from the blood of suckling rats more slowly than from that of the maternal lactating rats. Based on these data, a potential risk to breast feeding infants is identified.

The effects of istradefylline administration in combination with levodopa and carbidopa as proposed clinically was assessed in repeat dose studies in rat and dog and in an EFD study in rabbit. The doses in the rabbit combination EFD study were based on a DRF study conducted in pregnant female Japanese white rabbits at doses of istradefylline up to 800 mg/kg in combination with 80/20 mg/kg of LD/CD. In groups given 600 and 800 mg/kg Istradefylline, significant maternal toxicity and mortality and embryo-foetal mortality was observed. External malformations were noted in all groups dosed in excess of 200 mg/kg of Istradefylline. 400 mg/kg was chosen as the top dose for use in the pivotal combination EFD study.

In the pivotal study, no mortality or treatment related clinical signs were observed in any treated dams. Maternal body weight gain and food consumption was lower in the high dose (400/80/20 istradefylline/L/C mg/kg/day) relative to control. Lower foetal body weights were noted in the 400/80/20, and 200/80/20 istradefylline/L/C mg/kg/day groups, with decreased foetal viability noted in the higher dose group. visceral malformations (adactyly, brachydactyly, ectrodactyly, absent claw, absent metacarpal, absent metatarsal, and absent phalanx) were higher in the 400/80/20 mg/kg combination treatment group compared to the vehicle control, Istradefylline alone or L/C alone treatment groups. Digit malformations (in line with those noted in the istradefylline alone administration study) were observed in 13 foetuses from 6 dams and 2 foetuses from one dam in 400 and 200 mg combination groups respectively. Increased incidence of membranous ventricular septum defects were also observed in the high dose combination group.

One foetus in the low dose group exhibited a number of malformations (including digit malformations as well as craniofacial abnormalities). As these findings were not in line with effects noted at other doses and in the dedicated istradefylline EFD study this was considered non-test article related. This is accepted (only 1 foetus of 125 in this group exhibited any visceral malformations with 3 of 125 exhibiting skeletal malformations).

Combination administration in rabbits was shown to be associated with an increased exposure to istradefylline relative to istradefylline alone. It is not clear if a similar PK interaction occurs clinically. The low dose was set as the NOAEL in this study resulting in a ≈ 1.2-fold exposure relative to predicted clinical exposures at the proposed high dose clinically.

Toxicokinetic data

Toxicokinetics (TK) data show evidence of accumulation following repeated dosing. Of note, no adverse brain findings in terms of neuronal damage, inflammation, haemorrhage or gliosis were reported. No evidence of any effect on ECG parameters was noted in any of the pivotal istradefylline dog studies.

Three dog studies examining the effect of treatment with istradefylline in combination with levodopa and carbidopa as proposed clinically on PK and toxicity were submitted. PK data acquired in the pivotal 13-week oral administration combination toxicity study (B-5759) were highly variable, but mean istradefylline exposures were in excess of those recorded following administration of istradefylline alone. There was an increase in submandibular gland weight in females in the combination group in this study but this was without histopathological correlates. No other new target organs were identified though decreased heart rate and increased QT interval were noted in mid and high dose combination groups. This was attributed to L/C treatment. This is accepted.

TK data were acquired in all pivotal repeat-dose toxicity studies. In rats, exposures in terms of AUC_{0-24} increased in a less than dose proportional manner with lower exposures at the end of treatment relative to those on day one indicating potential metabolic induction. As discussed above, sampling time points from the 26-week repeat-dose toxicity study did not allow for the calculation of an AUC. Data from these studies were variable, with a sex related difference (lower exposures in females) more pronounced in the 4-week study utilising non-micronised istradefylline (a-95-133-94-210), than the 4-week study utilising micronised istradefylline (khk33-960498-khk-33). As expected, higher dose normalised exposures were evident in animals treated with micronized relative to non-micronised drug substance.

In dogs, exposures increased in a less than dose proportional manner. In contrast to rat studies there was evidence of accumulation with increased exposures evident following repeat dosing. Again data obtained were variable, with decreased exposures in females evident in some dose levels in some studies but not consistently.

The applicant has presented exposure margin calculations based on exposures observed in repeat dose rat and dog studies relative to those observed at the 20 and 40 mg/day doses clinically. In rats, exposure margins at the proposed higher clinical dose relative to no observed adverse effect level(s) (NOAELs) defined in pivotal studies were less than 1 for females and just over 1 for males. As noted above, no AUC data are available for the 26-week rat study and therefore exposure data from the 4-week study is used. In dogs, exposure margins at the higher dose relative to NOAELS defined in 26 and 52-week studies were less than 1 for both sexes (there was no significant sex related differences in exposure in dogs). NOAELS were defined due to adverse effects noted in the liver, kidney, adrenals, pancreas and lung at higher dose levels. In general, these effects were relatively minor and the applicant contends that the available safety data from clinical studies and post marketing experience do not indicate any significant human toxicities associated with these findings. The lack of safety margins to NOAELs identified in non-clinical studies should be taken in the context of the limitations in the clinical safety database and justification for the proposed higher dose.

It should be noted that in the 26-week rat study adverse effects above the NOAEL included foci of vascular mineralisation in the brain with similar findings observed in 2-year carcinogenicity studies in both rat and mouse. The applicant has provided a separate justification/risk assessment related to this finding.

Local Tolerance

No studies were provided by the applicant.

Other toxicity studies

Antigenicity

Studies to assess the potential direct or passive antigenicity of istradefylline have been conducted in rats, mice and guinea pigs and do not suggest any cause for concern.

Dependency

Potential physical dependence of istradefylline was assessed following 4 weeks of administration in food to rats. Animals behaviour and body weight were monitored over the course of a 7-day withdrawal period with diazepam administration in food acting as a positive control. The diazepam treated group exhibited a marked withdrawal syndrome evidenced by reduced food consumption and body weight gain. In contrast, the data presented do not suggest a withdrawal syndrome in istradefylline treated groups although reduced food consumption was noted in the low dose group on day 1 and 2 of withdrawal relative to controls. The applicant contends this was a rebound effect due to higher food consumption during treatment, this seems unlikely as total daily food intake was similar to control on day 1. However, as there is no dose response for this finding it is accepted that the data do not suggest a strong withdrawal syndrome.

A second study in rats examined the potential for physical dependence following twice daily dosing via oral gavage of two doses of istradefylline with morphine used as a positive control. Istradefylline treatment was associated with increased locomotor activity, with a higher proportion of rats exhibiting this on day 1 of treatment in both dose groups. This returned to baseline in the withdrawal phase. Istradefylline treatment was also associated with a dose dependent increase in food consumption. This dropped below control levels following cessation of treatment. Again, this was interpreted as a rebound effect rather than withdrawal. There was a small, non-statistically significant but consistent reduction in body weight following cessation of treatment at both dose levels utilised in this study. There was a significant decrease in food consumption and body weight in the morphine treated group following cessation of treatment. PK taken in this study show relevant exposures were 1.24 and 5.97 those anticipated clinically at the high dose in the low and high dose istradefylline groups respectively. Given that increased food consumption and body weight gain have been noted with Istradefylline in repeat dose toxicity studies in rat, these are not considered sensitive endpoints for assessment of dependency and these data are considered equivocal.

The potential reinforcing effect of istradefylline administration was assessed in a self-administration study in rhesus monkeys. This study reported an increase in self-administration of istradefylline relative to vehicle in 2 of 4 monkeys. However, the dimthylacetamidde/polyethylene glycol 400/saline vehicle used in this study was subsequently shown to exhibit significant toxicity in rhesus monkeys following exposures of 0.5ml to 1 ml/kg, similar to those used in the self-administration study at higher doses. This confounds the results of this study. Furthermore, each single IV dose produced exposures significantly lower than those anticipated at either dose proposed clinically, therefore the results of this study are not considered reliable.

In a d-amphetamine cued drug discrimination test in rats, istradefylline partially generalised to the d-amphetamine cue. Although this was not fully dose dependent, results were consistently higher than saline. Generalisability was to a significantly lesser extent than the clear dose proportional generalisation evident with d-amphetamine and phentermine, but was similar to that of bupropion.

It is accepted that the presented data do not indicate that Istradefylline is likely to induce significant physical dependence or has stimulant properties comparable to amphetamine. The study assessing reinforcing characteristics of Istradefylline was flawed and therefore these data are not considered

relevant. There are significant limitations in the non-clinical assessment provided placing greater weight on the results of the clinical dependence study conducted.

Impurities

The istradefylline cis-isomer (KF23546) was assessed for acute toxicity in rat up to doses of 2000 mg/kg and in a GLP complaint Ames test up to concentration of 2000 µg per plate due to precipitates formed at higher concentrations. No fatalities were recorded in the acute toxicity study and KF23546 was negative for mutagenicity in the Ames test. A similar approach was taken for istradefylline dimer (KF66275) with on mortality noted up to highest dose tested and negative for mutagenicity. KF66275 was also shown to have a relatively low affinity for the A_{2A} receptor.

Phototoxicity

The potential for Istradefylline to induce phototoxicity was assessed in an *in vitro* 3T3 fibroblast study and two *in vivo* studies in Long-Evans pigmented and RORO-n hairless rats. Istradefylline was reported as non-phototoxic up to a concentration of 14 µg/ml in cultured murine fibroblasts, higher concentrations could not be tested due to a lack of solubility. In RORO-n hairless rats, mild erythema was observed following exposures to UVA light $\geq$ 30 J/cm2 following either single or repeat administration of 400 mg/kg istradefylline via oral gavage. There was an increased incidence of this finding in animals administered the test article for 7 days. No similar reactions were observed in animals administered vehicle control.

In contrast, in a three-day test article administration study in long-Evans rats in which animals were dosed up to 400 mg/kg via oral gavage and exposed to UVR at a dose of 0.5 MED equivalent to approximately 100 J/M2, no treatment related effects on ophthalmological evaluations or skin reactions were observed. It is accepted that the totality of the data suggests that istradefylline is unlikely to induce phototoxicity at normal environmental light intensities.

Effects on adrenal related hormones

As an increase in adrenal weight and hypertrophy of cells in the zona fasciculata of the adrenals were the most consistent findings reported in repeat dose toxicity studies in both dog and rat, the applicant has submitted additional (non-GLP) mechanistic studies examining the effects of istradefylline administration on adrenal function. Istradefylline (it is not clear if this was micronized or non-micronized drug substance) administration in single doses as low as 25 mg/kg to SD rats resulted in an increase in plasma ACTH, corticosterone and aldosterone 4-hours post administration. This was not evident following repeated oral administration for 7-14 days suggesting an attenuation of this response following repetitive dosing. The applicant discussed the possible effect of istradefylline exposure on adrenal glands but the mechanism underlying hypertrophy of the zona fasciculata (cortex) observed in all repeat-dose studies in rats and dogs (but not in mice) and possible gender effect (female rats appeared to be consistently more sensitive), remains unclear.

Although the role of A_{2A} receptors in the hypothalamus-pituitary-adrenal axis is not completely clear, the possible interference by istradefylline on glucocorticoid secretion by adrenal zona fasciculata involving ACTH, cannot be excluded.

However, considering that the effects in animals were reversible, were not accompanied by functional (e.g., hormonal) alterations, did not include degeneration or necrosis in rat carcinogenicity studies, and no adrenal adverse drug events were evident from clinical trials, the potential safety risk in humans is expected to be low.

In dogs, there were no consistent changes in cortisol, aldosterone, insulin (this was elevated in one male animal) following 4 weeks treatment with micronized Istradefylline at a dose of 400 mg/kg. Note, no assessment of acute effects following a single dose was made in this study.

Effects on urinalysis and calcium homeostasis

An additional set of mechanistic studies was conducted to assess the effects of Istradefylline on urinalysis and calcium homeostasis. Istradefylline was shown to exhibit a diuretic effect in rats and mice in line with findings in the repeat dose toxicity study. Similar effects were not observed in dog. Increased levels of renin activity and plasma aldosterone were observed 3 hours following single administration of Istradefylline to rats but were not observed following 14 days administration. Increased urinary excretion of sodium, potassium, chloride, and calcium was observed from day one of 320 mg/kg treatment and following 2 weeks of treatment at 30 mg/kg. Istradefylline was also shown to increase urinary calcium excretion in SD rats at doses in excess of 10 mg/kg. This correlated with decreased bone mass in the high dose group. The applicant has submitted an independent review of these data which point out limitations in the study design and that interpretation of these data is difficult and suggest this is likely related to effects on growth rather than bone loss. This is accepted.

2.3.3. Ecotoxicity/environmental risk assessment

The submitted environmental risk assessment (ERA) was written by an appropriately qualified expert and is signed and date 20/7/20.

Log P was assessed using the shake-flask method (study d-04-502) and ranged from 3.5 to 3.6 and no Persistence, Bioaccumulation, Toxicity (PBT) screen was triggered. Although the document is signed by the 'quality study director', there is no GLP compliance statement with this study which is not in line with the EMAs 'Guideline on the environmental risk assessment of medicinal products for human use' (Doc. Ref. EMEA/CHMP/SWP/4447/00 corr 21*). The applicant subsequently confirmed it was compliant with the "Criteria for Reliability of Application Data (Article 43 of the Enforcement Regulations, Pharmaceutical Affairs Law)" of Japan which mandates that studies are conducted reliably and that accurate documentation are recorded. In the absence of a direct comparison between the differences between these two standards, the applicant's argumentation that this is unlikely to affect the quality or interpretation of the data generated in this study, while plausible, is unsubstantiated.

The applicant has confirmed that the study (d-04-502) was not conducted in accordance with OECD 107, specifically they note divergence in terms of the number of volume ratio conditions, the preparation method of saturated solutions, and the method of phase separation. They argue that the test results were consistent across runs and that the test system is robust.

It is considered that the divergence in the protocol to produce saturated solutions, the lack of full rotation of the test solution, the lack of centrifugation to separate the test solutions and, significantly, the lack of any discussion under what temperature the experiment was conducted could have a meaningful impact of the data generated in this experiment. While it is accepted that this is unlikely to dramatically alter the calculated Log P, in order to complete the ERA and accurately inform the ERA table in the European Public Assessment Report (EPAR). The applicant has committed to conduct an appropriate GLP compliant study to assess Log Kow in line with OECD 107 post authorisation.

Adsorption was characterised in 3 soil and 2 sludge types via a GLP-compliant OECD 106 study (Study LX42MM). Although several deviations to the study protocol were noted, these were not considered to adversely affect the interpretation of the data in this study and this is endorsed. Istradefylline was not readily biodegradable in an GLP compliant OECD 301B test (Study DT67SN) and an OECD 308 (Study RT73KN) aerobic transformation in water-sediment system showed a significant shift into sediment which triggered a sediment organism (OECD 218) toxicity assessment. The cis-isomer of istradefylline was the primary transformation product reported, with several other low-level degradants reported (≤8.6% in the total systems). Since isomers are not distinguished in the environmental risk assessment of pharmaceuticals the applicant was asked to sum up the radioactivities of istradefylline and its cis-isomer

and to recalculate the DT50 values for the water phases. Considering the cis-isomer, calculations lead to a DT50 of 127.2 days for total system 1 (Calwich Abbey Lake) at 12 °C via HS kinetics (best visual fit, Chi2 error = 5.7%) and thus, istradefylline has to be classified as persistent.

The applicant argued that as the NER and BR are unlikely to be bioavailable which is the focus of the ERA risk assessment, that the dissipation rates for the total system are appropriate for the assessment of persistence. They state that the majority of the NER were associated with humin and note that the conducted OECD 218 sediment organism toxicity study did not indicate a significant risk to sediment dwelling organisms.

However, humin matter fractionation is not an appropriate method to differentiate the various types of NER and to identify the fraction of irreversibly bound residues (see ECHA NER discussion paper, Kästner et al. 2018). According to ECHA R.11, 2017 NER should be considered as non-degraded substance in the absence of a systematic methodology, unless it can be analytically proven that a certain part can be considered as irreversibly bound.

The assessment of persistence must be made on the basis of degradation rather than dissipation half-lives. Therefore, on the basis of the available data, (considering the DT50 of 127.2 days in combination with the very high proportion of NER (88.1 % at end of the study) and the very low rate of mineralisation (< 1 %)), Istradefylline must be considered very persistent and the ERA table in the EPAR will reflect this. Therefore, due to the very high proportion of NER at the end of the test (70.3 %AR and 88.1 %AR), the DT50 values of the sediments and total systems should be set to > 1000 d and istradefylline should be classified as very persistent (vP).

Toxicity studies with aquatic organisms were conducted including growth inhibition in Raphidocelis subcapitata (OECD 201-Study HQ62QJ), reproduction in Daphnia magna (OECD 211-Study LQ23FW) and fish early-life stage toxicity in Pimephales promelas (OECD 210-Study NP53PP). Wet weight in Pimephales promelas in the fish early life stage toxicity study was the most sensitive endpoint with a NOEC of 1.5 µg/L. Concerning microorganisms in STP activated sludge Istradefylline did not exhibit significant toxicity at any of the concentrations tested in an activated sludge respiration inhibition test (OECD 209-Study GV91SY). Validity criteria were met for all studies, some minor protocol deviations were noted, but it is accepted these were unlikely to affect the outcomes of the studies in question and results are considered valid. Following the application of the appropriate assessment factors, PEC/PNEC comparisons do not indicate istradefylline as posing a threat to the environment.

As LogKow was > 3 a fish bioaccumulation study should be carried out in Phase II Tier B. The applicant has submitted an exploratory study demonstrating that istradefylline is not stable enough to perform an aqueous flow though bioaccumulation study. Although it is noted that Istradefylline is not superlipophilic, nor is solubility as low as suggested in the relevant guidance ((log Kow > 5 with solubility below ~0.01 – 0.1 mg/L), it is accepted that based on limitations in the solubility of Istradefylline, a dietary exposure bioaccumulation study in fish is appropriate. The applicant has conducted a GLP compliant dietary exposure bioaccumulation study in rainbow trout. Fish were exposed to [14C]-labelled Istradefylline at a concentration of 100 µg/g in feed over a 14 day period (Study 8427603). The validity criteria for the test were met. Following 14 days exposure, mean concentration of total radioactivity in fish tissue was 0.439 µg equivalents/g, declined to 0.020 µg equivalents/g (reduction of 95.4%) at Day 14 of depuration. Kinetic analysis estimated the biomagnification factor (BMFK) to be 0.00439 and the fish lipid normalized biomagnification factor corrected for growth rate (BMFKgL) to be 0.0292 (18% lipid food content) and 0.0325 (20% lipid food content). The growth corrected half-life (t1/2g) was estimated to be 0.5 days, suggesting a rapid elimination of radioactivity from fish tissues following a 14-day exposure to food treated with istradefylline. The applicant has applied a number of methods sourced from OECD 305 technical guidance document to estimate BCF from the derived BMF values summarised above. This

approach returned a range of values (range 165.3 to 776 Kg/L), one of which was in excess of the B trigger of 2000 Kg/L.

In general, it is accepted that this is an outlier and based on the totality of the data presented it can be concluded that istradefylline is not bioaccumulative. However, several of these derivations rely of the Log kow value of Istradefylline. The applicant has committed to resubmit the BCF calculations based on the Log-Kow derived from the requested OECD 107 GLP-compliant study.

The submitted OECD 218 study utilising Chironomus riparius met its validity criteria (with on minor deviation, which it is accepted was unlikely to impact on the interpretation of the data generated) and is considered acceptable. PECsediment was calculated using formula from the ECHA guidance document and PECsediment/PNECsediment was below 1 indicating an acceptable risk to sediment dwelling organisms.

Table 1: Summary of main study results

Substance (INN/Invented Name): Istradefylline			
CAS-number (if available): 155270-99-8			
PBT screening		Result	**Conclusion**
Bioaccumulation potential- log K_{ow}	Non GLP, Non-OECD 107 shake flask	3.5	Potential PBT (N) The applicant has been requested to submit an appropriately conducted study
PBT-assessment			
Parameter	**Result relevant for conclusion**		**Conclusion**
Bioaccumulation	log K_{ow}	3.5	Current BCF values based on non-acceptable logkow value and to be amended (unlikely to affect B assessment)
	BCF	165.3 to 776 kg/L	
Persistence	DT50 or ready biodegradability	Not readily biodegradable DT50, whole system >1000 days based on % shifting to sediment = 45.3 + 27.5 NER @ day 14	vP based on very high proportion of NER at the end of the test
Toxicity	NOEC Algae	850 µg /L	T toxic based on aquatic toxicity data
	NOEC Crustacea	66 µg/L	
	NOEC Fish	1.5 µg/L	
PBT-statement :	The compound is considered very Persistent and Toxic but not bioaccumulative		
Phase I			
Calculation	**Value**	**Unit**	**Conclusion**
PEC $_{surfacewater}$, refined based on EU disease prevalence	0.065	µg/L	> 0.01 threshold (Y)
Other concerns (e.g. chemical class)			(Y/N)
Phase II Physical-chemical properties and fate			
Study type	**Test protocol**	**Results**	**Remarks**
Adsorption-Desorption	OECD 106	Sludge K_{oc} = 354 and 887 Soil K_{oc} = 1750 21,100 565	2 sludge types, 3 soil types. Sludge <10000 L/Kg, no risk assessment for

					terrestrial compartment
Ready Biodegradability Test	OECD 301 B	Not readily biodegradable			
Aerobic and Anaerobic Transformation in Aquatic Sediment systems	OECD 308	Calwich Abbey Lake $DT_{50, water (12 ºC)}$ = 4.29 days $DT_{50, sediment (12 ºC)}$= > 1000 days $DT_{50, whole system (12 ºC)}$ > 1000 days % shifting to sediment = 25.1 + 48.8 NER @ day 14 Emperor Lake $DT_{50, water (12 ºC)}$ = 2.72 days $DT_{50, sediment (12 ºC)}$ > 1000 days $DT_{50, whole system (12 ºC)}$ > 1000 days % shifting to sediment = 45.3 + 27.5 NER @ day 14			Istradefylline + cis Isomer Dissipation rate for Calwich Abbey Lake DT50, whole system (12 ºC)= 127.2 days % partitioning to sediment >10%, hence Phase IIb sediment toxicity study triggered
Phase IIa Effect studies					
Study type	**Test protocol**	**Endpoint**	**value**	**Unit**	**Remarks**
Algae, Growth Inhibition Test/ *Raphidocelis subcapitata*	OECD 201	NOEC	850	µg /L	
Daphnia sp. Reproduction Test	OECD 211	NOEC	66	µg/L	
Fish, Early Life Stage Toxicity Test/*Pimephales promelas*	OECD 210	NOEC	1.5	µg/L	NOEC based on wet weight
Activated Sludge, Respiration Inhibition Test	OECD 209	NOEC	1 x 10^6	µg/L	
Phase IIb Studies					
Bioaccumulation	OECD 305	BCF	165.3 to 776	L/kg	Dietary administration study. BMFK = 0.00439 BMFKgL = 0.0292 (18% lipid food content) = 0.0325 (20% lipid food content) t1/2g was estimated to be 0.5 days
Sediment dwelling organism *(Chironomus riparius)*	OECD218	NOEC	851 4052	mg/ kg	o.c. content 2.1% normalized to 10% o.c. content

2.3.4. Discussion on non-clinical aspects

Pharmacology

The pharmacology package demonstrates that istradefylline is a potent and selective ligand of the adenosine A_{2A} receptor. Although, results from binding affinity assays demonstrate that in dogs istradefylline presented the lowest affinity for A_{2A} receptors (Ki = 65 nmol/L vs human 12.4 nmol/L and rats 8.18 nmol/L) and the lowest selectivity for A_{2A} over A_1 receptor (3.4-fold in dog, vs >80-fold in human, and 14-fold in rat). The applicant justified the choice of dog as a non-clinical species on the basis that monkeys presented a low absorption of istradefylline and there is precedent for the use of dogs in

these types of studies. The cardiovascular safety assessment of istradefylline in dogs was considered valid as the istradefylline concentrations achieved in the plasma dogs were above that required for inhibition of adenosine A_{2A} receptors, although it's expected that significant inhibition of A_1 receptors also occurred in dogs, whereas significant inhibition of A_1 receptors in humans at the usual clinical doses of 20 mg/day or 40 mg/day is unlikely. In addition, the absence of brain vascular mineralisation in dogs was considered unlikely to be due to insufficient A_{2A} antagonism, the applicant indicates that animals were sufficiently exposed to istradefylline to achieve significant adenosine A_{2A} antagonism.

The scientific rationale for the use of istradefylline in the proposed indication is based on the presence of adenosine A_{2A} receptors on neurons of the indirect pathway from the striatum to the globus pallidus, but not on neurons of the direct striate-pallidal pathway. These pathways input into the motor thalamus and in turn the motor cortex, with the direct pathway providing excitatory input and the indirect pathway decreasing excitatory input. Both pathways have dopaminergic receptors and both are impacted by dopaminergic depletion in PD, therefore the rationale for use of istradefylline in PD is to block activation of the inhibitory indirect pathway, by blocking activation of adenosine A_{2A} receptors, thereby reducing inhibitory input to the thalamus and motor cortex. The mode of action of istradefylline is via competitive inhibition of A_{2A} receptors and istradefylline was shown to inhibit adenosine A_{2A} receptor-mediated enhancement of GABAergic synaptic transmission in the GP in rats. Istradefylline was also shown to increase spontaneous locomotor activity in D2-/- mice, demonstrating this pharmacological effect is independent of D2 receptors which co-localise with A_{2A} receptors on indirect striate-pallidal pathway neurons. Proof-of concept studies completed in rat PD models indicate istradefylline potently antagonised haloperidol-induced catalepsy, without evidence of tolerance to the inhibitory effect of istradefylline for up to 14-days. Inhibitory activity beyond 14 days was not examined in any proof of concept study and notably hyperactivity findings in the rat repeat-dose toxicology studies resolve after approximately 2 weeks of istradefylline administration. Istradefylline was shown to reverse motor disability in MPTP treated marmosets in a dose-dependent manner for 21 days (Kanda, 1998, study #98-216), but evidence of a durability of effect of istradefylline beyond 21 days in non-clinical species is not available. However, the durability of effect of istradefylline is addressed in the clinical efficacy assessment and therefore, further non-clinical data was not requested.

Istradefylline, M1 and M4 were shown to have similar binding affinities for rat A_{2A} receptor, but M4 has a greater affinity than both M1 and istradefylline for rat A_{2A} receptors. A similar potency of M1 and istradefylline in the haloperidol-induced catalepsy model in mice was also demonstrated. However, the metabolic profile of istradefylline in rats and dogs was only qualitatively but non-quantitatively similar to that observed in humans. It appears that the level of M1 present in human plasma is low and there is a higher content of M5 and M8 in human plasma. Therefore, these data provide an incomplete picture of the potential contribution of the metabolites to the pharmacodynamics. Although the binding affinities of the metabolites M4, M5 and M8 to human A_{2A} receptors are unknown, the contribution of these metabolites to the observed pharmacodynamics activity of istradefylline is considered likely to be low. This is based on reported exposure levels for each of the metabolites in question, following 14 days of istradefylline dosing in humans, which are < 10% of the exposure to the parent drug (Study 6002-US-016). This is acceptable and further non-clinical characterisation of these metabolites are not required.

Secondary PD studies focussed on the cardiovascular effects of istradefylline but studies evaluating CNS, inflammatory effects and potential platelet aggregation effects were also included. No findings of note with regard to safety are reported, although an anti-inflammatory effect of istradefylline in a carageenan-induced inflammation model of unknown relevance is described.

Safety pharmacology studies indicate a significant CNS response of increased spontaneous locomotor activity following oral administration of istradefylline in rodents and common marmosets. Additionally, shortened pentobarbital-Na-induced sleeping time occurred in mice and increased rectal temperatures occurred in mice, rats and rabbits. Cardiovascular safety pharmacology studies indicate increased heart

rate and blood pressure in conscious rats following oral istradefylline administration but this effect was likely the result of an increase in spontaneous locomotor activity and it was not replicated in conscious dog studies, although locomotor activity levels were not reported. A finding of reduced femoral blood flow in anaesthetised dogs is of uncertain relevance and the applicant suggests it may be a result of the experimental preparation. Given the potential effect of Istradefylline towards adenosine receptors located in skeletal muscle and endothelial smooth muscle cells and the finding of myocyte vacuolation observed in mouse carcinogenicity studies from 250 mg/kg, the applicant was asked to better discuss the role of Istradefylline in skeletal muscular tissue and its possible clinical consequences. The applicant acknowledged the reductions in skeletal muscle blood flow and increases in femoral vascular resistance encountered in Study KHK25a-960333 but did not consider the findings reliable as only single doses were tested, the dogs were under general anaesthesia, and the gradual deterioration of the experimental preparation is likely to have led to cooling and reduced metabolic demand in the skeletal muscle of the leg (resulting in reduced femoral blood flow and an increase in calculated vascular resistance). On the other hand, the myocyte vacuolation observed in carcinogenicity studies following long-term exposure to istradefylline appears not to be associated with an inflammatory response or other types of lesions. Therefore, although the mechanism of the effects of istradefylline on rodent skeletal muscle remains unknown, it is acknowledged that from a clinical point of view, no safety signals concerning skeletal muscle were observed, therefore, it's plausible to believe that istradefylline presents a low safety risk to the skeletal muscle function in humans.

In general, pivotal safety pharmacology studies were conducted in compliance with GLP but the *in vitro* hERG assay is non-GLP compliant. The applicant does not consider the lack of GLP-compliance to have impacted upon study endpoints in the hERG assay as the study is compliant with the "Criteria for Reliability of Application Data (Article 43 of the Enforcement Regulations, Pharmaceutical Affairs Law)" of Japan, which reportedly ensures studies are performed reliably and reproducibly, with appropriate records of protocol compliance, verification and retention of data. This issue was not pursued further.

Pharmacokinetics

Single dose absorption studies were conducted several species, including rats, dogs and rabbits, relevant species for the toxicology studies. In rats a dose-proportional increases in systemic exposure in rats occurred up to 30mg/kg, at which point exposure was saturated and oral bioavailability reduced from approximately 20%, to 7% at 100mg/kg. Exposure in rabbits at 100mg/kg was considerably higher than in rats, with mortality occurring at the 300mg/kg dose level in rabbits. Istradefylline exposure increased less than dose-proportionally in dogs and bioavailability reduced from over 100% at 0.3mg/kg, to 19% at 100mg/kg. Istradefylline was shown to be absorbed throughout the gastrointestinal tract in rats, with absorption in the duodenum> small intestine > rectum > colon > stomach.

Both male and female animals were included in repeat-dose toxicokinetic studies conducted on the 4-week toxicology studies in rats and dogs, allowing for the assessment of gender-based differences in PK. The sponsor has also included studies using both micronized and un-micronized istradefylline in rats allowing for comparison. Istradefylline did not accumulate during repeated dosing in male and female rats but, accumulation did occur with repeat dosing in dogs. No gender-related differences are reported in either species. In general, exposures to micronized istradefylline at doses of 100 mg/kg and 400 mg/kg were similar to that obtained in the non-micronized istradefylline study at doses of 160 mg/kg and 800 mg/kg respectively, indicating that micronizing istradefylline increased exposure as anticipated. The study reports from 4-week repeat-dose toxicology studies in rats (#KHK33/963444) and dogs (#A-96-83), following daily oral administration of istradefylline include GLP-compliance statements but the toxicokinetics are included in separate study reports (#A-96-139 and #A-96-29 respectively) which are non-GLP compliant. The sponsor does not consider the lack of GLP-compliance to have impacted upon study endpoints or the conclusions drawn from the toxicology studies on the basis that the final study

reports were audited by the Kyowa Kirin Co., Ltd. Quality Assurance Unit at its GLP facility and considered to have accurately described the test method and results. This issue is not pursued further.

Tissue distribution and Foetal Transfer studies in pregnant rats (#96-644A, 10591 #96-643A) indicate significant foetal exposure to istradefylline and/ or metabolites, which is higher on GD19 than GD12 suggesting that placental transfer of istradefylline occurred more easily at the later stage of pregnancy than during organogenesis.

Toxicology

The applicant has submitted an extensive safety package for Istradefylline as expected for a product with such a long development. The primary findings from the repeat-dose toxicity studies identify pancreas (cell vacuolation and apoptosis), lung (increased alveolar macrophages) adrenal glands (hypertrophy of the zona fasciculata), liver (centrilobular hyperthrophy associated to liver enzymes, total/direct bilirubin increase), kidney (vacuolation in the epithelia of the proximal tubuli), and brain (vascular mineralisation). Exposure margins to clinical AUC reached at mg/day are quite low and not all safety findings can be attributed to the exaggerated pharmacological activity. The applicant has justified this by referencing the extensive clinical safety database for these drugs. It is accepted that this is a clinical issue and no further justification from a non-clinical perspective has been sought.

There are significant limitations in the PK data available from pivotal toxicity studies. This was specifically problematic for the rat EFD studies where appropriate TK data from pregnant rats are available. It was not considered that requesting new TK data from pregnant rats at this stage will be informative due to the historic nature of EFD studies. However, CHMP has no grounds to question the validity of the EFD study data either.

Dose and treatment-duration related foci of brain vasculature mineralisation were observed following chronic administration in rat repeat dose toxicity and carcinogenicity study. Similar findings were not observed following chronic administration in dog and similar findings observed in the mouse carcinogenicity study were age related with similar incidence in treated and control groups. The applicant has provided a separate justification document related to this finding outlining why this is not considered clinically relevant. They have conducted a full review of all pivotal toxicity studies and it is accepted that further non-clinical studies are unlikely to add further to our current understanding of the clinical relevance of this risk. The data is not considered sufficient to conclude on the clinical relevance of this risk.

2.3.5. Conclusion on the non-clinical aspects

An extensive non-clinical package has been presented in support of this marketing authorisation application. There are no non-clinical issues outstanding which preclude the authorisation of Nouryant.

2.4. Clinical aspects

2.4.1. Introduction

GCP

The Clinical trials were performed in accordance with GCP as claimed by the applicant.

The applicant has provided a statement to the effect that clinical trials conducted outside the Community were carried out in accordance with the ethical standards of Directive 2001/20/EC.

- Tabular overview of clinical studies

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Biopharmaceutic Studies **Bioavailability (BA) Study Reports**								
Phase 1 Food effect	6002-011	Effect of food on PK of istradefylline; safety of istradefylline	Randomized, OL, single-center, 2-period crossover study	Istradefylline 20 mg tablet, oral *Group A*: fed → fasted *Group B*: fasted → fed	*Overall*: 20M 29 (20 to 34) years *Group A*: 10M 28 (20 to 34) years *Group B*: 10M 29 (22 to 33) years	Non-smoking, healthy Japanese subjects	Single doses separated by ≥21-day washout period	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Biopharmaceutic Studies (continued) **BA Study Reports (continued)**								
Phase 1 Safety, PK, BA	6002-9601	Safety and single-dose PK of istradefylline; effect of a meal on BA of istradefylline	*Steps 1 to 6*: Randomized, SB, PC, parallel-group study *Step 7*: 2-period, 2-sequence crossover study	Istradefylline 10, 25, or 50 mg tablet, oral; *Steps 1 to 6*: 10, 25, 50, 100, 150, or 200 mg istradefylline, or placebo, single doses under fasted conditions; *Step 7*: 50 mg single dose under fasted and fed conditions	60M NC (20 to 29 years) 48 istradefylline 12 placebo *Step 1*: 22 (20 to 24) years *Step 2*: 23 (20 to 24) years *Step 3*: 23 (22 to 24) years *Step 4*: 22 (20 to 25) years *Step 5*: 24 (21 to 27) years *Step 6*: 23 (20 to 28) years *Step 7*: 24 (21 to 29) years	Healthy Japanese subjects	*Steps 1 to 6:* Single doses *Step 7:* Single doses separated by a 2-week washout period	Completed; Final
Phase 1 Food effect	6002-US-023	Effect of a high-fat meal on BA of istradefylline	Randomized, OL, single-center, 2-period crossover study	Istradefylline 40 mg tablet, oral; 40 mg single dose; *Sequence 1*: fed → fasted *Sequence 2*: fasted → fed	27M 24 (18 to 46) years	Non-smoking, healthy subjects	Single doses separated by 22-day washout period	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Biopharmaceutic Studies (continued) **Comparative BA and Bioequivalence (BE) Study Reports**								
Phase 1 BE	6002-012	BE of 2 istradefylline 10 mg tablets compared to one 20 mg tablet	Randomized, OL, single-center, 2-group, 2-period, crossover study	*Group A*: Day 1: Istradefylline 10 mg (x2) tablet, oral Day 13: Istradefylline 20 mg tablet, oral *Group B*: Day 1: Istradefylline 20 mg tablet, oral Day 13: Istradefylline 10 mg (x2) tablet, oral	*Overall*: 30M 26 (20 to 37) years *Group A*: 15M 26 (20 to 37) years *Group B*: 15M 27 (21 to 34) years	Non-smoking, healthy Japanese subjects	Single doses separated by ≥12-day washout period	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Biopharmaceutic Studies (continued) **Comparative BA and BE Study Reports (continued)**								
Phase 1 BE	6002-US-022	*Cohort 1*: Intra-subject variability of istradefylline PK *Cohort 2*: Bioequivalence of istradefylline 40 mg tablets from lots used in clinical studies compared to the 40 mg intended commercial tablet	*Cohort 1*: NR, OL, uncontrolled, single-center, 2-period, replicate study *Cohort 2*: Randomized, OL, single-center, 3-period, 6-sequence, crossover study	*Cohort 1*: Istradefylline 40 mg intended commercial tablet, oral; 40 mg single dose on Days 1 and 22 *Cohort 2*: Istradefylline 40 mg tablets, oral; A: formulation used in Phase 2b studies B: formulation used in Phase 3 studies C: intended commercial tablet 40 mg single dose on Days 1, 22, and 43	*Cohort 1*: 14M 34 (24 to 43) years *Cohort 2*: 96M 33 (18 to 45) years	Non-smoking, healthy subjects	Single doses separated by 21-day washout period	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human Pharmacokinetic (PK) Studies **Healthy Subject PK and Initial Tolerability Study Reports**								
Phase 1 Safety, PK	6002-EU01	Safety, tolerability, and single-dose PK of istradefylline	Randomized, DB, PC, single-center, ascending single-dose, crossover study	Istradefylline 5, 10, 25, 50, and 100 mg capsules, oral; *Group 1*: Single ascending doses of 5, 25, 100, and 300 mg istradefylline, or placebo; *Group 2*: Single ascending doses of 10, 50, 200, and 400 mg istradefylline, or placebo	18M 31 (22 to 53) years 18 istradefylline 16 placebo (*Note: 2 subjects discontinued and were replaced*)	Healthy subjects	Single doses separated by at least 14-day washout period	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Healthy Subject PK and Initial Tolerability Study Reports (continued)**								
Phase 1 Safety, PK, BA	6002-9601	Safety and single-dose PK of istradefylline; effect of a meal on BA of istradefylline	*Steps 1 to 6*: Randomized, SB, PC, parallel-group study *Step 7*: 2-period, 2-sequence crossover study	Istradefylline 10, 25, or 50 mg tablet, oral; *Steps 1 to 6*: 10, 25, 50, 100, 150, or 200 mg istradefylline, or placebo, single doses under fasted conditions; *Step 7*: 50 mg single dose under fasted and fed conditions	60M NC (20 to 29 years) 48 istradefylline 12 placebo *Step 1*: 22 (20 to 24) years *Step 2*: 23 (20 to 24) years *Step 3*: 23 (22 to 24) years *Step 4*: 22 (20 to 25) years *Step 5*: 24 (21 to 27) years *Step 6*: 23 (20 to 28) years *Step 7*: 24 (21 to 29) years	Healthy Japanese subjects	*Steps 1 to 6:* Single doses *Step 7:* Single doses separated by a 2-week washout period	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Healthy Subject PK and Initial Tolerability Study Reports (continued)**								
Phase 1 Safety, PK	6002-EU02	Safety, tolerability, and repeated-dose PK of istradefylline (pilot study)	Randomized DB, PC, multiple-dose, single-center, parallel-group study	Istradefylline 25 and 50 mg capsules, oral; *Group 1:* 25 mg/day istradefylline or placebo for 14 days; *Group 2:* 75 mg/day istradefylline or placebo for 14 days[a]	12: 9M/3F 31 (22 to 39) years 9 istradefylline 3 placebo	Healthy subjects	14 days	Completed; Final
Phase 1 Safety, PK	6002-9703	Safety and repeated-dose PK of istradefylline	Randomized, SB, PC multiple-dose, single-center, parallel-group study	Istradefylline 20-mg tablet, oral; 20 mg/day istradefylline or placebo for 14 days	12M 27 (21 to 30) years 9 istradefylline 3 placebo	Non-smoking, healthy Japanese subjects	14 days	Completed; Final
Phase 1 Safety, PK	6002-0104	Safety and repeated-dose PK of istradefylline	Randomized, SB, PC, single-center, sequential group, multiple-dose, ascending dose study	Istradefylline 20-mg tablet, oral; *Group 1:* 20 mg/day istradefylline or placebo for 14 days; *Group 2:* 40 mg/day istradefylline or placebo for 14 days; *Group 3:* 80 mg/day istradefylline or placebo for 14 days	36M 24 (20 to 42) years 27 istradefylline 9 placebo	Non-smoking, healthy Japanese subjects	14 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Healthy Subject PK and Initial Tolerability Study Reports (continued)**								
Phase 1 Safety, PK	6002-US-002	Safety, tolerability, and repeated-dose PK of istradefylline	Randomized, third-party blinded, PC, single-center, sequential group, multiple-dose, ascending dose study	Istradefylline 20 mg tablet, oral; *Group 1:* 40 mg/day istradefylline or placebo for 14 days; *Group 2:* 60 mg/day istradefylline or placebo for 14 days; *Group 3:* 80 mg/day istradefylline or placebo for 14 days; *Group 4:* 120 mg/day istradefylline or placebo for 14 days; *Group 5:* 160 mg/day istradefylline or placebo for 14 days	50M 33 (21 to 45) years 40 istradefylline 10 placebo	Healthy subjects	14 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Healthy Subject PK and Initial Tolerability Study Reports (continued)**								
Phase 1 Safety, PK	PP15710	Safety, tolerability, and multiple-dose PK of istradefylline, including investigation of possible effects of gender and smoking on PK	Randomized, DB, PC, parallel-group, ascending multiple-dose study	Istradefylline 10 mg capsule, oral; *Group 1*: 10 mg/day istradefylline or placebo for 14 days; *Group 2*: 20 mg/day istradefylline or placebo for 14 days; *Group 3*: 40 mg/day istradefylline or placebo for 14 days; *Group 4*: 40 mg/day istradefylline or placebo for 14 days	56: 32M/24F NC (55 to 77 years) 42 istradefylline 14 placebo 10-mg Group: 6M/6F 63 (55 to 75) years 20-mg Group: 6M/6F 59 (55 to 68) years 40-mg Group: 6M/6F 59 (55 to 77) years 40-mg (smokers) 6M Group: 61 (57 to 67) years Placebo Group: 6M/6F 60 (55 to 69) years Placebo (smokers) Group: 2M 65 (58 to 72) years	*Groups 1 to 3*: Non-smoking, healthy elderly subjects *Group 4*: Smoking, healthy elderly subjects	14 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Healthy Subject PK and Initial Tolerability Study Reports (continued)**								
Phase 1 QT/QTc, PK, Safety	6002-US-024	Effect of istradefylline on ECG (thorough QTc study); PK and safety at therapeutic and supratherapeutic doses	Randomized, DB, PC, single-center, parallel-group study, with moxifloxacin (SB) active control	Istradefylline 40 mg tablets, oral; Moxifloxacin 400 mg tablets, oral *Group 1*: 40 mg/day istradefylline for 14 days; *Group 2*: 160 mg/day istradefylline for 14 days; *Group 3*: 400 mg/day moxifloxacin on Days 1 and 14, placebo on Days 2 to 13; *Group 4*: placebo for 14 days	176: 88M/88F 33 (18 to 45) years Group 1: 44: 22M/22F 32 (18 to 44) years Group 2: 44: 22M/22F 33 (19 to 45) years Group 3: 44: 22M/22F 34 (18 to 45) years Group 4: 44: 22M/22F 32 (19 to 45) years	Non-smoking, healthy subjects	14 days	Completed; Final
Phase 1 PK, metabolism, excretion	6002-US-010	Single-dose PK, metabolism, and excretion of istradefylline	NR, OL, single-dose, mass-balance study	Oral suspension or capsule containing approx. 100 µCi [^{14}C]-istradefylline; single dose of approx. 40 mg	9M 28 (18 to 53) years	Healthy subjects	Single doses	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Patient PK and Initial Tolerability Study Reports**								
Phase 1 Safety, PK	6002-US-003[b]	Safety and repeated-dose PK of istradefylline	NR, OL, uncontrolled, sequential group, multiple ascending-dose study	Istradefylline 20 mg tablet, oral; *Group 1*: 60 mg/day istradefylline for 14 days; *Group 2*: 80 mg/day istradefylline for 14 days	10: 8M/2F 60 (48 to 76) years	Patients with Parkinson's disease under primary treatment with levodopa/carbidopa	14 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Intrinsic Factor PK Study Reports**								
Phase 1 Safety, PK	6002-EU03	Safety and single-dose PK of istradefylline	Randomized, DB, PC, uncontrolled, 3-period crossover, single rising-dose study	Istradefylline 50 and 100 mg capsules, oral; Single doses of 50, 100, and 150 mg istradefylline or placebo	19: 9M/10F 68 (60 to 79) years 19 istradefylline 18 placebo (*Note: 1 replacement subject was recruited after 1 subject withdrew*)	Healthy elderly subjects	Single doses separated by 28-day washout period	Completed; Final
Phase 1 Safety, PK	6002-0205	Safety and single-dose PK of istradefylline	NR, OL, uncontrolled, single-dose study	Istradefylline 20 mg tablet, oral; Single dose of 40 mg	18M *Group A (elderly)*: 73 (67 to 82) years *Group B (non-elderly)*: 24 (20 to 33) years	Healthy Japanese subjects	Single doses	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Intrinsic Factor PK Study Reports (continued)**								
Phase 1 PK, Safety	6002-US-015	Effect of severe renal impairment on single-dose PK and safety of istradefylline	NR, OL, multicenter, parallel-group study	Istradefylline 40 mg tablet, oral; 40 mg single dose *Group 1*: Renally impaired subjects *Group 2*: Healthy subjects matched to Group 1 by age, BMI, and gender *Group 3*: Healthy young subjects	*Overall*: 18: 12M/6F 41 (20 to 69) years *Group 1*: 6 (4M/2F) 52 (37 to 69) years *Group 2*: 6 (4M/2F) 48 (37 to 61) years *Group 3*: 6 (4M/2F) 23 (20 to 28) years	Subjects with severe renal impairment (creatinine clearance <30 mL/min) and healthy subjects	Single dose	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Intrinsic Factor PK Study Reports (continued)**								
Phase 1 PK, Safety	6002-016	Effect of mild hepatic impairment on single-dose PK of istradefylline and selected metabolites; safety and tolerability of istradefylline	NR, OL, two center, one-sequence, two-period, parallel-group crossover study	Istradefylline 40 mg tablet, oral	*Overall*: 20: 10M/10F 54 (33 to 69) years *Mild Hepatic Impairment:* 10: 5M/5F 52 (33 to 65) years *Normal Hepatic Function*: 10: 5M/5F 55 (40 to 69) years	Non-smoking subjects, healthy or with mild hepatic impairment (Child-Pugh Grade "A", score 5-6 inclusive) matched for age, sex, race and BMI	Single dose	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Intrinsic Factor PK Study Reports (continued)**								
Phase 1 PK, Safety	6002-US-016	Effect of moderate hepatic impairment and smoking on repeated-dose PK of istradefylline and selected metabolites; safety of istradefylline	NR, OL, multicenter, parallel-group study	Istradefylline 40 mg tablet, oral; 40 mg/day for 14 days; *Group 1*: smokers with moderate hepatic impairment; *Group 2*: healthy smokers, matched to Group 1 by age, BMI, and gender; *Group 3*: non-smokers with moderate hepatic impairment; *Group 4*: healthy non-smokers, matched to Group 3 by age, BMI, and gender	28: 16M/12F 57 (45 to 77) years *Group 1:* 7: 5M/2F 57 (49 to 65) years *Group 2*: 7: 5M/2F 53 (45 to 64) years *Group 3*: 7: 3M/4F 60 (47 to 77) years *Group 4*: 7: 3M/4F 56 (45 to 73) years	Smoking (at least 20 cigarettes per day) and non-smoking subjects, healthy or with moderate hepatic impairment (Child-Pugh Grade "B", score 7-9 inclusive)	14 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Extrinsic Factor PK Study Reports**								
Phase 1 PK (drug-drug interaction study)	BP15748	Effect of istradefylline on PK of levodopa/ carbidopa	Randomized, DB, PC, single-center, multiple-dose study	Substrate: *Group 1 and 2:* Levodopa/carbidopa 100/25 mg t.i.d. for 21 days (Days 1 to 21), oral; Interacting Drug: Istradefylline, 20 mg capsule, oral *Group 1*: 20 mg/day istradefylline or placebo for 14 days (Days 8 to 21); *Group 2*: 40 mg/day istradefylline or placebo for 14 days (Days 8 to 21)	32: 16M/16F NC (18 to 38) years 24 istradefylline 8 placebo	Healthy subjects	21 days	Completed; Final
Phase 1 PK (drug-drug interaction study)	6002-US-009	Effect of istradefylline on PK of levodopa/ carbidopa	NR, OL, single-center, fixed sequence study	Substrate: Levodopa/carbidopa 100/25 mg tablets, oral; 200/50 mg single dose on Days 1 and 15 Interacting Drug: Istradefylline 40 mg tablet, oral; 80 mg/day for 14 days (Days 2 to 15)	24: 17M/7F 36 (24 to 49) years	Healthy subjects	15 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Extrinsic Factor PK Study Reports (continued)**								
Phase 1 PK (drug-drug interaction study)	BP15809	Effect of istradefylline on PK of midazolam	NR, OL, single-center, multiple-dose, sequential-group study	Substrate: Group 1 and 2: Midazolam 7.5 mg single dose on Days 1 and 16, oral; Interacting Drug: Istradefylline 5 and 20 mg tablets, oral; Group 1: 5 mg/day for 14 days (Days 3 to 16) Group 2: 20 mg/day for 14 days (Days 3 to 16)	25M NC (21 to 44) years	Non-smoking, healthy subjects	16 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Extrinsic Factor PK Study Reports (continued)**								
Phase 1 PK (drug-drug interaction study)	6002-US-008	Part 1: Effect of istradefylline on PK of midazolam Part 2: Effect of ketoconazole on PK of istradefylline	NR, OL, single-center, multiple-dose, sequential, 2-period study	Part 1: Substrate: Midazolam 10 mg single dose on Days 1 and 17, oral; Interacting Drug: Istradefylline 40 mg tablet, oral; 80 mg/day for 15 days (Days 4 to 18) Part 2: Substrate: Istradefylline 40 mg tablet, oral; 40 mg single dose on Days 1 and 19; Interacting Drug: Ketoconazole 200 mg b.i.d. for 4 days (Days 15 to 18) and 200 mg q.d. for 7 days (Days 19 to 25), oral	Part 1: 17: 13M/4F 36 (18 to 54) years Part2: 18: 13M/5F 29 (20 to 44) years	Healthy subjects	Part 1: 18 days Part2: 25 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Extrinsic Factor PK Study Reports (continued)**								
Phase 1 PK (drug-drug interaction study)	6002-US-020	Effect of istradefylline on PK of atorvastatin	Randomized, DB, PC, single-center, fixed-sequence study	Substrate: Atorvastatin 40 mg single doses on Days 1 and 18, oral; Interacting Drug: Istradefylline 40 mg tablet, oral; 40 mg/day istradefylline or placebo for 17 days (Days 5 to 21)	20M 16 istradefylline 44 (21 to 54) years 4 placebo 39 (28 to 48) years	Healthy subjects	21 days	Completed; Final
Phase 1 Food effect	6002-US-023	Effect of a high-fat meal on BA of istradefylline	Randomized, OL, single-center, 2-period crossover study	Istradefylline 40 mg tablet, oral; 40 mg single dose; *Sequence 1*: fed → fasted *Sequence 2*: fasted → fed	27M 24 (18 to 46) years	Non-smoking, healthy subjects	Single doses separated by 22-day washout period	Completed; Final
Phase 1 Food effect	6002-011	Effect of food on PK of istradefylline; safety of istradefylline	Randomized, OL, single-center, 2-period crossover study	Istradefylline 20 mg tablet, oral *Group A*: fed → fasted *Group B*: fasted → fed	*Overall*: 20M 29 (20 to 34) years *Group A*: 10M 28 (20 to 34) years *Group B*: 10M 29 (22 to 33) years	Non-smoking, healthy Japanese subjects	Single doses separated by ≥21-day washout period	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Extrinsic Factor PK Study Reports (continued)**								
Phase 1 Safety, PK, BA	6002-9601	Safety and single-dose PK of istradefylline; effect of a meal on BA of istradefylline	*Steps 1 to 6*: Randomized, SB, PC, parallel-group study *Step 7*: 2-period, 2-sequence crossover study	Istradefylline 10, 25, or 50 mg tablet, oral; *Steps 1 to 6*: 10, 25, 50, 100, 150, or 200 mg istradefylline, or placebo, single doses under fasted conditions; *Step 7*: 50 mg single dose under fasted and fed conditions	60M NC (20 to 29 years) 48 istradefylline 12 placebo *Step 1*: 22 (20 to 24) years *Step 2*: 23 (20 to 24) years *Step 3*: 23 (22 to 24) years *Step 4*: 22 (20 to 25) years *Step 5*: 24 (21 to 27) years *Step 6*: 23 (20 to 28) years *Step 7*: 24 (21 to 29) years	Healthy Japanese subjects	*Steps 1 to 6:* Single doses *Step 7:* Single doses separated by a 2-week washout period	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Extrinsic Factor PK Study Reports (continued)**								
Phase 1 PK (drug-drug interaction study)	6002-015	Effect of rifampin on PK of istradefylline	NR, OL, single-center, 1-sequence, 2-period study	*Period 1*: Istradefylline 40 mg single dose on Day 1 tablet, oral *Period 2*: Substrate: Istradefylline 40 mg single dose on Day 8 tablet, oral; Interacting Drug: Rifampin 600 mg (2 x 300 mg capsules) for 20 days capsules, oral	20: 14M/6F 46 (34 to 65) years	Non-smoking, healthy subjects	20 days	Completed; Final
Phase 1 PK (drug-drug interaction study)	6002-US-026	Effect of istradefylline on PK of digoxin	NR, OL, single-center, fixed sequence study	Substrate: Digoxin 0.4 mg (2 x 0.2 mg capsules) single dose on Days 1 and 35, oral; Interacting Drug: Istradefylline 40 mg tablet for 21 days (Days 15 to 35), oral	24M 28 (20 to 39) years	Non-smoking, healthy subjects	21 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PK Studies (continued) **Extrinsic Factor PK Study Reports (continued)**								
Phase 1 PK, Safety	6002-US-016	Effect of moderate hepatic impairment and smoking on repeated-dose PK of istradefylline and selected metabolites; safety of istradefylline	NR, OL, multicenter, parallel-group study	Istradefylline 40 mg tablet, oral; 40 mg/day for 14 days; *Group 1*: smokers with moderate hepatic impairment; *Group 2*: healthy smokers, matched to Group 1 by age, BMI, and gender; *Group 3*: non-smokers with moderate hepatic impairment; *Group 4*: healthy non-smokers, matched to Group 3 by age, BMI, and gender	28: 16M/12F 57 (45 to 77) years *Group 1:* 7: 5M/2F 57 (49 to 65) years *Group 2*: 7: 5M/2F 53 (45 to 64) years *Group 3*: 7: 3M/4F 60 (47 to 77) years *Group 4*: 7: 3M/4F 56 (45 to 73) years	Smoking (at least 20 cigarettes per day) and non-smoking subjects, healthy or with moderate hepatic impairment (Child-Pugh Grade "B", score 7-9 inclusive)	14 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human Pharmacodynamic (PD) Studies **Healthy Subject PD and PK/PD Study Reports**								
Phase 1 PD, PK, Safety	6002-EU06	Adenosine A_{2A} receptor occupancy of istradefylline in the human brain (PET study); PK and safety of istradefylline	NR, OL, uncontrolled, multicenter, multiple-dose, parallel-group study	Istradefylline (unlabelled) 0.1 and 0.5 mg capsules, or 5 and 20 mg tablets, oral; [^{11}C]-istradefylline (300 MBq in 5 to 10 mL ethanol solution), i.v. injection; *Group 1:* single i.v. dose of [^{11}C]-istradefylline on Day 0; *Groups 2 to 8:* 40, 20, 5, 1.5, 0.5, or 0.1 mg/day istradefylline for 14 days (Days -13 to 0) + single i.v. dose [^{11}C]-istradefylline on Day 0	15M[c] 51 (37 to 64) years 13 istradefylline 2 [^{11}C]-istradefylline	Healthy subjects	14 days	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Human PD Studies (continued) **Healthy Subject PD and PK/PD Study Reports (continued)**								
Phase 1 Abuse Potential, Safety	6002-017	Abuse potential, safety, and tolerability of istradefylline in recreational stimulant users	Randomized, DB, placebo- and active-controlled, single-center, 6-way crossover study	Qualification Phase: X: Phentermine 60 mg, oral Y: Placebo, oral Treatment Phase: A: Istradefylline 40 mg, oral B: Istradefylline 80 mg, oral C: Istradefylline 160 mg, oral D: Phentermine 45 mg, oral E: Phentermine 90 mg, oral F: Placebo, oral	Qualification Phase: 94 Treatment Phase: 55; 40M/15F 27 (19 to 44) Completed All Treatment Phases: 26 (19 to 38)	Healthy subjects with a history of recreational stimulant use	Single doses of each treatment. Washouts: 48 hours between X and Y 4 days between qualification and treatment 21 days each between A through F	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Efficacy and Safety Studies **Study Reports of Controlled Clinical Studies Pertinent to the Claimed Indication**								
Phase 2 Efficacy, Safety, PK	6002-EU04[e]	Efficacy, safety, and PK of istradefylline in de novo patients with Parkinson's disease	Randomized, DB[f], PC, multicenter, parallel-group, ascending multiple dose study	Istradefylline 5, 10, or 20 mg capsules, oral; 5, 10, and 20 mg/day istradefylline for 14 days each (ascending-dose design) or placebo for 42 days	19: 12M/7F 14 istradefylline 57 (37 to 75) years 5 placebo 60 (55 to 63) years	Untreated patients with idiopathic Parkinson's disease	6 weeks	Completed; Final
Phase 2 Efficacy, Safety, PK, PD	6002-EU05	Efficacy, safety, and PK, and effect of istradefylline on dyskinesia in patients with Parkinson's disease	NR, OL, uncontrolled, single-center, single ascending-dose, pilot study	Istradefylline 50 mg capsules, oral; Single doses of 50, 100, 200, and 300 mg on the first day of Weeks 0, 2, 4, and 6	10: 8M/2F 57 (43 to 66) years	Patients with Parkinson's disease	Up to 4 single doses, each separated by 2-week intervals	Completed; Final
Phase 2a Efficacy, Safety, PK	6002-US-001[d]	Efficacy, safety and PK of istradefylline as adjunctive therapy to levodopa/carbidopa in patients with Parkinson's disease	Randomized, DB, PC, parallel-group, multicenter, ascending multiple-dose study	Istradefylline 5, 10, and 20 mg tablets, oral; *Group 1*: 5/10/20 mg/day istradefylline for 28 days per dose *Group 2*: 10/20/40 mg/day istradefylline for 28 days per dose *Group 3*: placebo for 12 weeks	83: 48M/35F 54 istradefylline 64 (40 to 78) years 29 placebo 61 (42 to 81) years	Patients with idiopathic Parkinson's disease treated with levodopa/ carbidopa	12 weeks (i.e., 3 periods of 4 weeks each)	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Efficacy and Safety Studies (continued) **Study Reports of Controlled Clinical Studies Pertinent to the Claimed Indication (continued)**								
Phase 2 Efficacy, Safety, PK	6002-US-004	Efficacy, safety, and PK of istradefylline in patients with Parkinson's disease	Randomized, DB, PC, parallel-group, single-center, ascending multiple-dose study	Istradefylline 20 mg capsules, oral; 40 mg/day istradefylline or placebo for 2 weeks, followed by 80 mg/day istradefylline or placebo for 2 weeks	15: 8M/7F 61 (42 to 78) years 12 istradefylline 3 placebo	Patients with Parkinson's disease treated with levodopa/ carbidopa	6 weeks (2-week placebo run-in, 4 weeks of dose-escalation)	Completed; Final
Phase 2b Efficacy, Safety, PK	6002-US-005[d]	Efficacy, safety and PK of istradefylline in patients with Parkinson's disease	Randomized, DB, PC, multicenter, fixed multiple-dose, parallel-group study	Istradefylline 40 mg tablets, oral; 40 mg/day istradefylline or placebo for 12 weeks	195: 117M/78F 63 (38 to 87) years 129 istradefylline 66 placebo	Patients with Parkinson's disease treated with levodopa/ carbidopa	12 weeks	Completed; Final
Phase 2b Efficacy, Safety, PK	6002-US-006[d]	Efficacy, safety, and PK of istradefylline in patients with Parkinson's disease	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 20 and 40 mg tablets, oral; *Group 1*: 20 mg/day istradefylline for 12 weeks; *Group 2*: 60 mg/day istradefylline for 12 weeks; *Group 3*: placebo for 12 weeks	395: 264M/131F 64 (36 to 87) years 318 istradefylline 77 placebo	Patients with Parkinson's disease treated with levodopa/ carbidopa	12 weeks	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Phase 3 Efficacy, Safety, PK	6002-US-013	Efficacy, safety, and PK of istradefylline in patients with Parkinson's disease	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 20 mg tablets, oral; 20 mg/day istradefylline or placebo for 12 weeks	230: 153M/77F 64 (36 to 87) years 115 istradefylline 115 placebo	Patients with Parkinson's disease treated with levodopa/ carbidopa	12 weeks	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Efficacy and Safety Studies (continued) **Study Reports of Controlled Clinical Studies Pertinent to the Claimed Indication (continued)**								
Phase 3 Efficacy, Safety, PK	6002-US-018	Efficacy, safety, and PK of istradefylline in patients with Parkinson's disease	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 10, 20, or 40 mg tablets, oral; 10, 20 and 40 mg/day istradefylline or placebo for 12 weeks	605: 403M/202F 63 (35 to 87) years 454 istradefylline 151 placebo	Patients with Parkinson's disease treated with levodopa/ carbidopa	12 weeks	Completed; Final
Phase 3 Efficacy, Safety, PK	6002-EU-007	Efficacy, safety, and PK of istradefylline in patients with Parkinson's disease	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 20 mg tablets, oral; 40 mg/day istradefylline, 200 mg/day entacapone, or placebo for 16 weeks	464: 284M/180F 159 istradefylline 61 (35 to 81) years 153 entacapone 61 (34 to 87) years 152 placebo 62 (39 to 80) years	Patients with Parkinson's disease treated with levodopa in combination with either benserazide or carbidopa	16 weeks	Completed; Final
Phase 2a Safety, Efficacy	6002-0406	Efficacy and safety of istradefylline as adjunctive therapy in patients with Parkinson's disease	Randomized, DB, PC, multicenter, parallel-group, exploratory study	Istradefylline 10 mg and 20 mg tablets, oral; 20 or 40 mg/day or placebo	89: 39M/50F Placebo: 65 years Istradefylline 20 mg: 63 years Istradefylline 40 mg: 65 years	Patients with Parkinson's disease treated with levodopa	12 weeks	Completed; Final
Phase 2 Efficacy, Safety	6002-US-051	Efficacy and safety of istradefylline as monotherapy in subjects with Parkinson's disease	Randomized, DB, PC, parallel-group study	Istradefylline 40 mg tablets, oral; 40 mg/day istradefylline or placebo for 12 weeks	176: 103M/73F 63 (33-88) years 94 istradefylline 82 placebo	Patients with Parkinson's disease	12 weeks	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Efficacy and Safety Studies (continued) **Study Reports of Controlled Clinical Studies Pertinent to the Claimed Indication (continued)**								
Phase 3 Efficacy, Safety, PK	6002-009	Efficacy, safety, and PK of istradefylline in patients with Parkinson's disease	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 10 (x2) or 20 mg (x2) tablets, oral; 20 and 40 mg/day istradefylline or placebo for 12 weeks	373: 166M/207F 66 (42 to 84) years 247 istradefylline 126 placebo	Patients with Parkinson's disease treated with levodopa/ carbidopa	12 weeks	Completed; Final
Phase 2b Efficacy, safety, and PK	6002-0608	Efficacy, safety, and PK of istradefylline in patients with Parkinson's disease	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 10 (x2) or 20 mg (x2) tablets, oral; 20 and 40 mg/day istradefylline or placebo for 12 weeks	362; 150M/212F 65 (37 to 84) 243 istradefylline 119 placebo	Patients with Parkinson's disease treated with levodopa/ carbidopa	12 weeks	Completed; Final
Phase 3 Efficacy, safety, and PK	6002-014	Efficacy, safety, and PK of istradefylline in patients with Parkinson's disease	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 20 or 40 mg/day istradefylline or placebo orally for 12 weeks	612: 375M/237F 64 (40 to 87) years 408 istradefylline 204 placebo	Patients with Parkinson's disease treated with levodopa in combination with either benserazide or carbidopa	12 weeks	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Efficacy and Safety Studies (continued) **Study Reports of Uncontrolled Clinical Studies**								
Phase 2b/3 Safety and Efficacy	6002-US-007[g]	Long-term tolerability, safety, and efficacy of istradefylline in patients with Parkinson's disease	NR, OL, uncontrolled, multicenter flexible-dose, study	Istradefylline 20 or 40 mg tablet, oral; *Starting dosage:* 40 mg/day for 2 weeks *Maintenance dosage:* 20, 40, or 60 mg/day for up to 50 weeks (dose at discretion of investigator)	496: 317M/179F 64 (36 to 87) years	Patients with Parkinson's disease treated with levodopa/ carbidopa, with previous participation in a prior double-blind istradefylline trial	52 weeks[g]	Completed; Final
Phase 3 Safety	6002-INT-001	Long-term tolerability and safety of istradefylline in patients with Parkinson's disease	NR, OL, uncontrolled, multicenter, flexible-dose, study	Istradefylline 20 mg tablet, oral; *Starting dosage:* 40 mg/day for 2 weeks; *Maintenance dosage:* 20 or 40 mg/day for up to 50 weeks (dose at discretion of investigator)	1241: 819M/422F 63 (35 to 87) years	Patients with Parkinson's disease who completed Studies 6002-US-013, US-018, or EU-007, or who had enrolled in Study 6002-US-007	52 weeks	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Efficacy and Safety Studies (continued) **Study Reports of Uncontrolled Clinical Studies (continued)**								
Phase 3 Long Term Safety	6002-US-025	Long-term tolerability and safety of istradefylline in subjects with Parkinson's disease	NR, OL, uncontrolled multicenter, flexible dose study	Istradefylline 20 mg tablet, Same as previous dosage (last 2 weeks in study 6002-INT-001): 20 mg/day istradefylline 40 mg/day istradefylline	503: 348M/155F 65 (36 to 88) years	Patients with Parkinson's disease who have completed Week 52 of Study 6002-INT-001	2 years	Completed; Final
Phase 3 Safety	6002-010	Long-term tolerability and safety of istradefylline in patients with Parkinson's disease	NR, OL, uncontrolled, multicenter, flexible-dose, study	Transition Phase: 20 or 40 mg/day istradefylline or placebo, orally for 1 to 4 weeks (same drug assignments as 6002-009) OL Phase: Dose Adjustment: 20 mg/day orally for 4 weeks; Maintenance: Dose increase to 40 mg/day permitted at Week 4 Dose decrease to 20 mg/day permitted at Week 8	307: 133M/174F 65 (33 to 84) years	Patients with Parkinson's disease who completed Study 6002-009	52 week (OL Phase)	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Efficacy and Safety Studies (continued) **Study Reports of Uncontrolled Clinical Studies (continued)**								
Phase 3 Safety and Efficacy	6002-018	Long-term tolerability, safety, and efficacy of istradefylline in patients with Parkinson's disease	NR, OL, uncontrolled, multicenter, flexible-dose, study	Istradefylline 20 or 40 mg tablet, oral; *Starting dosage:* 20 mg/day for 12 weeks *Maintenance dosage:* 20 or 60 mg/day for up to 40 weeks (dose at discretion of investigator)	239: 152M/87F 65 (40 to 86) years	Patients with Parkinson's disease treated with levodopa/ carbidopa who completed Study 6002-014	52 weeks	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Efficacy and Safety Studies (continued) **Other Clinical Study Reports**								
Phase 2 Efficacy, Safety (other indication)	6002-US-101	Efficacy and safety of istradefylline in patients with Major Depressive Disorder	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 60 mg capsules (containing 3 tablets of 20 mg), oral; 120 mg/day istradefylline or placebo for 6 weeks	118: 46M/72F 41 (18 to 62) years 59 istradefylline 59 placebo	Patients with Major Depressive Disorder	6 weeks, preceded by 1-week placebo run-in	Completed; Final
Phase 2 Efficacy, Safety (other indication)	6002-US-104	Efficacy and safety of istradefylline in patients with Major Depressive Disorder	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 20 mg tablets, oral; *Group 1:* 40 mg/day istradefylline or placebo for 6 weeks; *Group 2:* 120 mg/day istradefylline for 6 weeks; *Group 3:* placebo for 6 weeks	243: 78M/165F 35 (18 to 69) years 163 istradefylline 80 placebo	Patients with single episode or recurrent Major Depressive Disorder	6 weeks, preceded by 1-week placebo run-in	Completed; Final
Phase 2 Safety, Efficacy (other indication)	6002-US-012[h]	Safety and potential therapeutic efficacy of istradefylline in patients with Restless Legs Syndrome	OL, uncontrolled study	Istradefylline 40 mg tablets, oral; 80 mg/day istradefylline for 6 weeks (planned treatment duration)	5F 62 (52 to 69) years	Patients with idiopathic Restless Legs Syndrome exhibiting sensory and motor abnormalities	6 weeks planned[h]	Completed; Final

Type of Study	Study Identifier	Objectives of the Study	Study Design and Type of Control	Test Product(s); Dosage Regimen; Route of Administration	Number of Subjects Treated M/F Mean Age (Range)	Healthy Subjects or Diagnosis of Patients	Duration of Treatment	Study Status; Type of Report
Reports of Efficacy and Safety Studies (continued) **Other Clinical Study Reports (continued)**								
Phase 2 Efficacy, Safety (other indication)	6002-US-201	Efficacy, safety, and tolerability of istradefylline in patients with Restless Legs Syndrome	Randomized, DB, PC, multicenter, fixed-multiple dose, parallel-group study	Istradefylline 40 mg tablets, oral; 40 mg/day istradefylline or placebo for 6 weeks	207: 99M/108F 54 (20 to 90) years 99 istradefylline 108 placebo	Patients with primary or idiopathic Restless Legs Syndrome	6 weeks	Completed; Final
Phase 2 Safety, Efficacy	6002-0407	Efficacy and safety of istradefylline as monotherapy in patients with Parkinson's disease	Randomized, DB, PC, multicenter, crossover study	Istradefylline 20 mg tablets, oral; 40 mg/day or placebo	72: 32M/40F 65 (30 to 85) years	Patients with Parkinson's disease	4 weeks/ treatment, separated by 4-week washout period	Completed; Final

a: 75 mg treatment arm of study cancelled.
b: Phase 1/2 study: completed in patients but safety/tolerability only (no efficacy endpoints).
c: Twelve completed, 10 of whom received unlabelled istradefylline for ≥ 14 days.
d: Patients had to be on levodopa/carbidopa and have both end of dose wearing-off and peak dose dyskinesias.
e: First administration in patients with Parkinson's disease.
f: Coordinating investigator remained unblinded in this multicenter study.
g: Study 6002-US-007 was prematurely discontinued by the Sponsor and only a few subjects had visits beyond 52 weeks.
h: Study 6002-US-012 was prematurely discontinued by the Sponsor.

BA=bioavailability, BE=bioequivalence, b.i.d=twice daily, BMI=body mass index, DB=double-blind, ECG=electrocardiogram, F=female, i.v.=intravenous, M=male, NC=not calculated, NR=non randomized, OL=open-label, PC=placebo-controlled, PET=positron emission tomography, PK=pharmacokinetics, SB=single-blind, t.i.d=three times a day

2.4.2. Pharmacokinetics

29 Phase 1 studies were conducted to evaluate the clinical PK of istradefylline including relative bioavailability, mass balance, DDI, effects of food, and special population studies. Istradefylline PK in patients with PD was assessed in one Phase 1 and 12 Phase 2/3 studies. Three population PK (pop-PK) analyses were conducted.

Analytical methods

All bioanalytical method validation reports, and associated data analysis reports, were assessed in the context of the latest EMA recommendations (EMEA/CHMP/EWP/192217/2009 Rev. 1 Corr. 2**).

Bioanalytical Method Validation

A total of 17 different bioanalytical methods were established throughout the clinical development programme to determine istradefylline, and associated metabolites, in various matrixes i.e. plasma, urine and faces. The methods were established across a range of sites, including Japan, Europe and the United States.

Istradefylline and associated metabolites were determined via high-performance liquid chromatography combined with UV detection (HPLC-UV), or HPLC combined with single quadrupole (LC-MS) or tandem mass spectrometric detection (LC/MS-MS).

HPLC-UV

Depending on the matrix (e.g. plasma, urine or faeces), istradefylline and associated metabolites were extracted from each sample following the addition of various solvents combined with centrifugation. Solvents used during various extraction phases included acetonitrile and ethyl acetate. Following reconstitution in an appropriate mobile phase, sample extracts were chromatographed on an HPLC column and quantified with UV detection.

LC-MS

Istradefylline was extracted from plasma samples using a mixture of ethyl acetate/n-hexane (7:3) combined with centrifugation. Following collection of the organic phase, and subsequent drying under nitrogen, samples were reconstituted with a suitable mobile phase prior to injection into the HPLC system. A single-quadrupole mass spectrometer was used as a detection system.

LC-MS/MS

Again, depending on the matrix, istradefylline and associated metabolites were extracted from each sample following the addition of different solvents, including, methyl t-butyl ether; or an ethyl acetate/n-hexane mixture. Extracts were subsequently chromatographed using HPLC and detected via MS/MS technologies.

Analytical Run Validation and Sample Analysis

During the analysis of participant samples, spiked CS and Quality Control (QC) samples were extracted to facilitate the accurate and precise determination of istradefylline, in addition to associated metabolites, in participant samples.

Samples were reanalysed if there were clear analytical grounds for rejection (e.g., interferences in the chromatogram, instrument failure, poor chromatography, faulty injection etc.). Incurred sample reanalysis (ICR) was not performed.

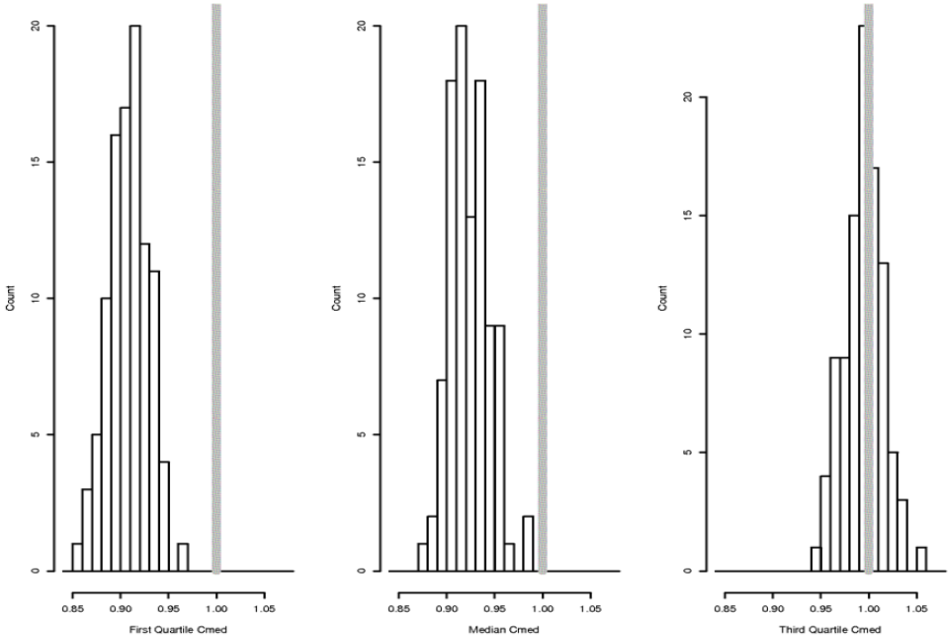
Population PK Analyses

6002-pop-pk-analysis (2006): Pop-PK analysis of istradefylline in healthy subjects and in patients with PD

Data from 8 Phase 1 and 8 Phase 2/3 clinical trials were included in this analysis. The database was comprised of 1449 subjects contributing a total of 10909 plasma istradefylline concentrations. There were 1219 PD patients and 230 healthy subjects.

The final popPK model was a two-compartment model with linear clearance and first-order absorption. Covariate effects included in the final model were Lean Body Mass (LBM), age, race, concomitant CYP3A4 inhibitors, and smoking status on oral clearance (CL/F); Weight on V2/F; LBM on Q/F; Weight on V3/F; and food status on F. The predictive check for the final model is presented below.

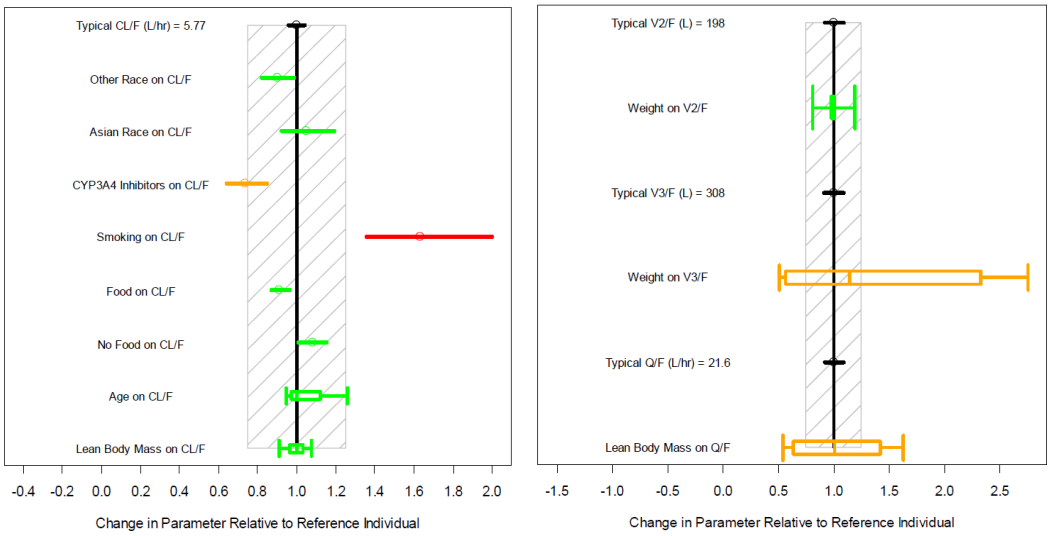
Figure 1: Predictive check for median istradefylline concentration



Simulated median concentrations plotted as histograms. The observed first quartile, median and third quartile median concentrations are indicated by a solid grey vertical line.

The main predictors of istradefylline exposure were smoking and the presence of CYP3A4 inhibitors as concomitant medications (Figure 2).

Figure 2: Effect of covariates on istradefylline pharmacokinetics.

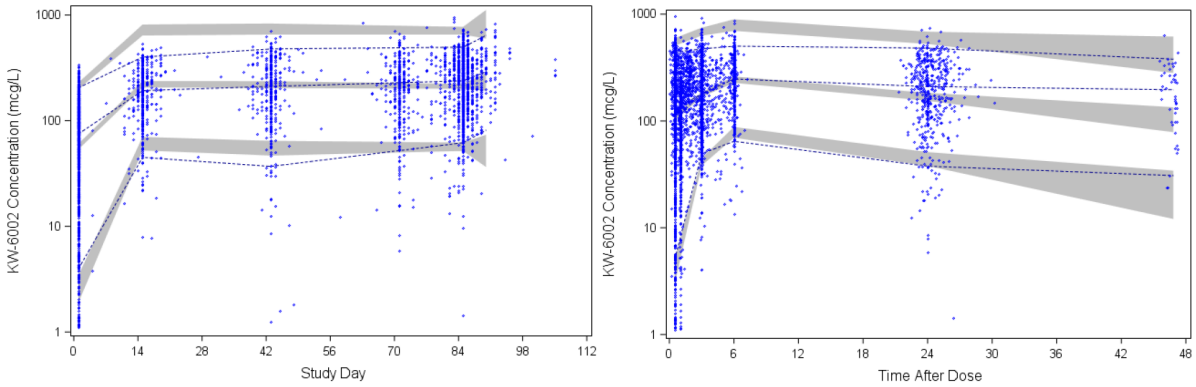


6002-014-pop-pk-r-en (2017): popPK analysis of istradefylline in patients with PD

This analysis utilised PK data collected in the single, multicenter Phase 3 study of istradefylline (Study 6002-014). The final database contained 3502 concentration records from 406 subjects.

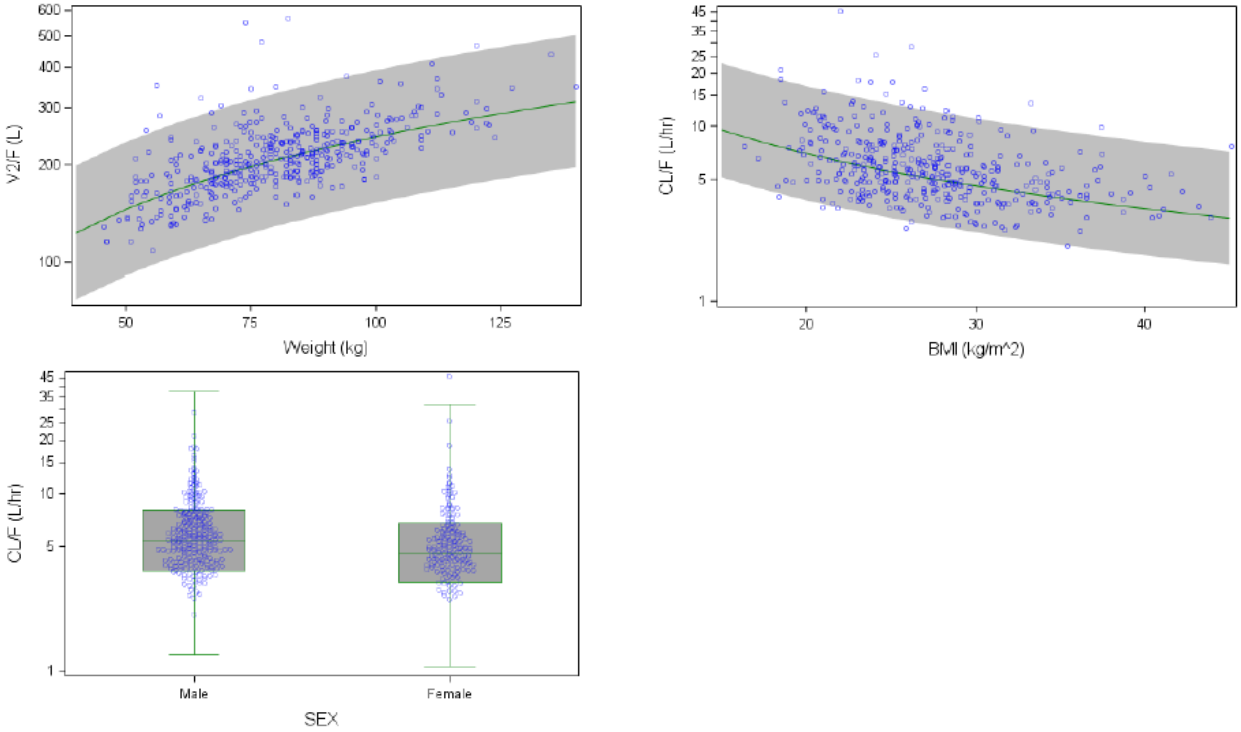
The final popPK model was a two-compartment model with linear clearance and three absorption transit compartments absorption. Covariate effects included in the final model were body mass index (BMI) and Sex on CL/F, and body weight on V1/F. The visual predictive checks (VPCs) for the final model are presented in Figure 3.

Figure 3: VPC results for final model – full profile (left) and Week 12 profile (right).



Simulations were conducted to examine the effect of identified covariates on popPK parameters (Figure *4*).

Figure 4: Effect of weight on V2/F (top left), BMI on CL/F (top right), and Sex on CL/F (bottom)

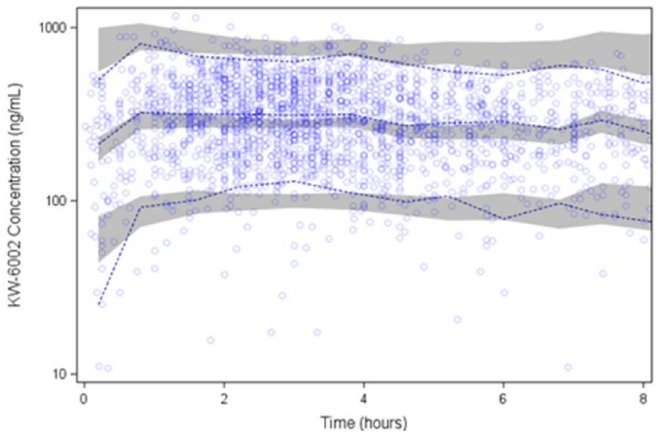


6002-nda-response-pop-pk-r-en (2018): popPK analysis of istradefylline in patients with PD

This popPK analysis was based on plasma concentration data from 5 phase 2/3 clinical studies: 6002-US-005, 6002-US-006, 6002-US-013, 6002-0608, and 6002-009. The final dataset included 2645 concentration records from 1034 subjects.

The final popPK model was a two-compartment model with linear clearance and first-order absorption. Covariate effects included in the final model were weight, Asian race, Sex and bilirubin on CL/F, and weight on V1/F. The VPC for the final model is presented in *Figure 5*.

Figure 5: VPC Results for Final Model-Full Profile



Simulations were conducted to examine the effect of identified covariates on popPK parameters relationship between popPK parameters (Figure 6 through Figure 10).

Figure 6

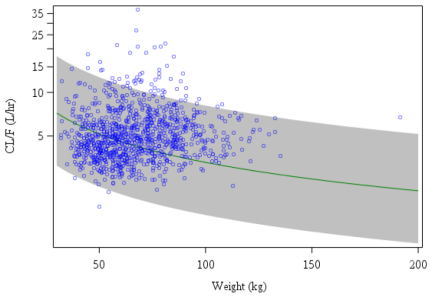


Figure 7

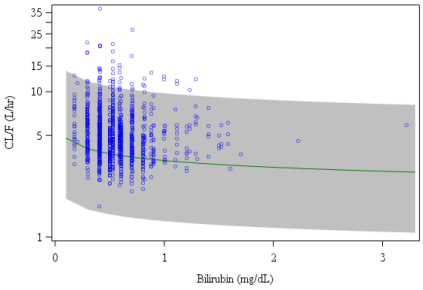


Figure 8

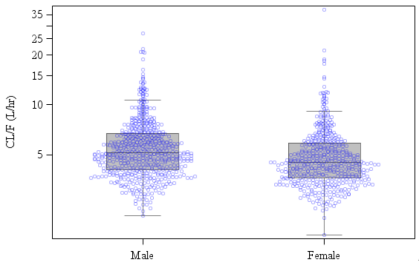


Figure 9

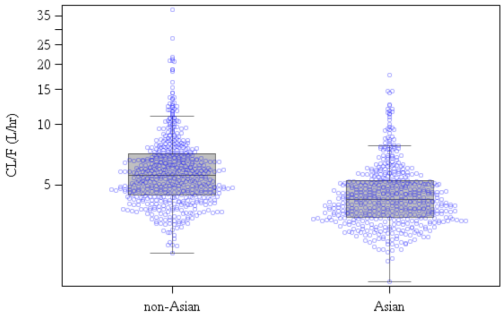
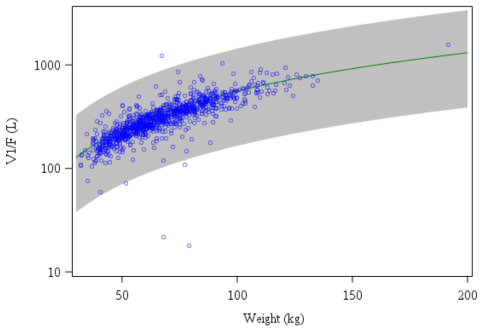


Figure 10



Absorption

Absorption

- **Bioavailability**

Single dose PK of istradefylline in healthy volunteers

Study 6002-9601 was a placebo-controlled, single-blind (SB) study investigating single ascending oral doses of istradefylline [10-200 mg] in healthy young adult male Japanese volunteers. PK parameters increased in a dose-dependent manner: C_{max} ranged from 43.0 to 342.3 ng/mL, $AUC_{0-\infty}$ ranged from 822.2 to 12240.6 (ng· h/mL) for 10-150 mg doses, t_{max} was observed at 2.5 to 3.5 hours.

Study 6002-EU01 was a single centre, randomized, double-blind (DB), placebo-controlled, 4-way cross-over study in healthy European male volunteers to investigate the PK of istradefylline following single ascending doses [5-400 mg]. Istradefylline C_{max} and AUC_{0-72} both increased in a dose dependent manner, ranging from 16.6 to 507.1 ng/mL and 302.1 to 19949 ng·h/mL, respectively. t_{max} was observed between 2 to 5 hours. $t_{1/2}$ ranged from 19.8 to 47.7 hours

Multiple dose PK of istradefylline in healthy volunteers

Study 6002-US-002 investigated the safety and PK characteristics of istradefylline following administration of multiple ascending doses [40-160 mg/day] for 14 days in healthy male volunteers. Steady state appeared to be achieved by Day 14 for doses up to 80 mg

Table 2: Summary Statistics for Selected PK Variables

Variable	Treatment Group Mean Value (SD):				
	40 mg/day (n=8)	60 mg/day (n=8)	80 mg/day (n=8)	120 mg/day (n=8)	160 mg/day (n=8)
C_{max} (ng/mL):					
Day 1:	282.4 (74.8)	399.9 (39.8)	430.4 (79.5)	495.1 (79.5)	678.6 (133.0)
Day 14:	598.7 (136.0)	719.9 (252.6)	955.5 (226.4)	1694.6 (330.5)	1974.9 (395.1)
T_{max} (h):					
Day 1:	2.38 (1.06)	3.13 (0.99)	3.38 (1.19)	3.00 (1.07)	3.00 (0.53)
Day 14:	3.38 (1.19)	3.75 (0.71)	3.25 (1.17)	4.88 (1.64)	3.63 (1.19)
$AUC_{0\text{-}24}$ (ng·hr/mL):					
Day 1:	2737.2 (774.8)	4274.5 (414.8)	5017.4 (797.0)	5830.5 (582.5)	7834.3 (1275.1)
Day 14:	9816.9 (2740.1)	12001.5 (4503.8)	16521.2 (5159.2)	31103.0 (6527.2)	36487.6 (6881.3)

Study 6002-EU02 was a DB, placebo controlled phase I trial in healthy male (n=6) and female (n=3) volunteers to investigate the PK of istradefylline and metabolite M1 following daily administration of a 25 mg tablet for 14 days.

Table 3: Mean (SD) PK Parameters derived from Plasma Concentrations of KW-6002 in Healthy Human Subjects administered single oral doses of 25mg/day for 14 days

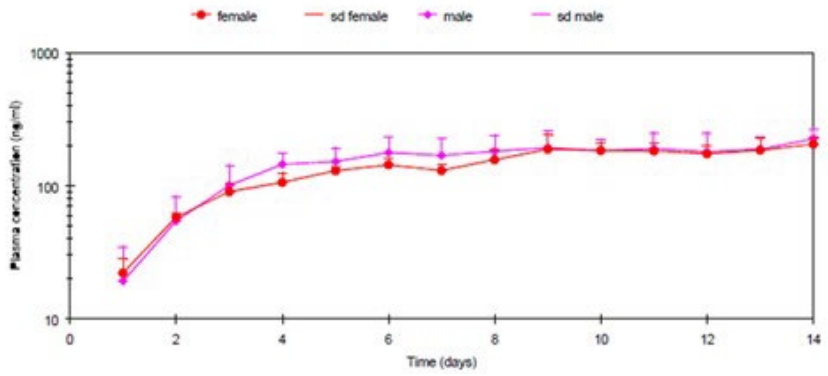
Day	C_{max} (sd) (ng/ml)	T_{max} (hours)	AUC_{24} (sd) (ng·h/ml)	λ_z (sd) ($hours^{-1}$)	$t_{½}$ (sd) (hours)
1	117.7 (23.2)	2[a]	1084.3 (248.6)	-	-
14	344.7 (61.1)	2[a]	5980.1 (927.1)	0.0214 (0.0176)	32.3[b]

a Value quoted is the median
b Calculated as ln2/mean λz

The mean terminal rate constant of istradefylline following the last dose on Day 14 of the study was 0.0214 hours-1, which corresponded to a $t_{1/2}$ of 32.3 hours (

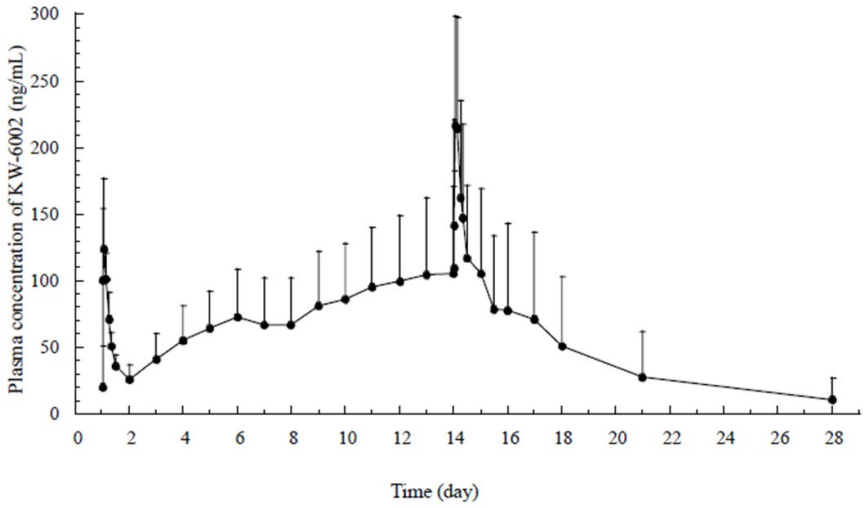
Table 3). However, the mean terminal rate constant in female subjects (n=3) was 0.0047 hours-1, corresponding to a $t_{1/2}$of 148.5 hours and the mean terminal rate constant in male subjects (n=6) was 0.0298 $hours^{-1}$, corresponding to a $t_{1/2}$ of 23.2 hours, which suggests a marked sex difference in the $t_{1/2}$ of istradefylline. Steady-state, however, was reached after approximately 8 days for both male and female participants (Figure 11), which places significant doubt on the accuracy of the terminal half-lives calculated.

Figure 11: Mean plasma KW-6002 concentrations at 24 Hours Post-Dose in Healthy Human Subjects administered single oral doses of 25mg KW-6002/Day for 14 days.



Study 6002-9703 was a placebo-controlled, SB, single-site study to investigate the safety and PK properties of istradefylline following repeated administration of one 20 mg tablet for 14 consecutive days in healthy Japanese adult male volunteers. The mean plasma trough concentrations (C_{24}) of istradefylline prior to the 12th, 13th, and final (14th) administration were 99.9 ± 48.8, 104.8 ± 57.2 and 105.6 ± 65.1, respectively. Moreover, C_{24} following the final administration was 105.0 ± 63.9 ng/mL, suggesting that steady state was approximately reached after 14 days of repeat dosing (Figure 12).

Figure 12: Mean plasma concentration of KW-6002 during and after multiple administration of KW-6002 at a dose of 20mg once a day for 14 days in healthy male volunteers.



Each symbol with a bar represents the mean +SD of nine volunteers.

Study 6002-0104 was a placebo-controlled, SB, ascending dose study to investigate the safety and PK properties of istradefylline in healthy Japanese adult male volunteers. Participants received 20, 40, or 80 mg of istradefylline, or placebo, once daily for 14 consecutive days. The plasma PK of istradefylline appeared to have reached steady state after 14 days of multiple dosing.

- **Bioequivalence**

Study 6002-US-022 was an open-label (OL), 3-period, 6-sequence, randomized, variance-balanced, orthogonal, Latin square study to investigate the bioequivalence of the final intended commercial formulation of istradefylline 40 mg oral tablets with alternative formulations used previously in Phase 2b (6002-US-005 and 6002-US-006) and Phase 3 (6002-US-018) clinical studies.

The least-squares mean (LSM) PK parameters with 90% confidence intervals (CI) for evaluation of bioequivalence are listed in Table 4. Bioequivalence was established between the final commercial formulation and alternative formulations used in previous clinical studies.

Table 4: LSM of PK parameters for the evaluation of bioequivalence (N=65)

PK Parameter	Treatment (40 mg tablet) A Batch F0946001	B Batch C3K0019	C Batch C4J0261	Ratio C/A (%)	90% CI	Ratio C/B (%)	90% CI
AUC_{0-t}	6655.51	6715.95	6685.21	100.45	(94.46, 106.81)	99.54	(93.61, 105.85)
$AUC_{0-\infty}$	7056.74	7110.27	7063.07	100.09	(94.03, 106.54)	99.31	(93.26, 105.81)
C_{max}	185.92	167.00	169.71	91.28	(85.95, 96.95)	101.62	(95.68, 107.93)
$t_{1/2}$	71.92	69.38	69.38	96.47	(91.33, 101.61)	99.33	(94.61, 105.38)

Study 6002-012 was a single-centre, 2-group, 2-period cross-over study to investigate the bioequivalence of 2 x 10 mg tablets compared to 1 x 20 mg tablet of istradefylline in healthy, non-smoking, Japanese males.

Differences in the mean for C_{max} and AUC_{0-t} after administration of each formulation (i.e. 10 and 20 mg tablets) were within the acceptable bioequivalence range of 80-125%

Table 5: Summary of plasma PK parameters

Formulation	Pharmacokinetic Parameter	Statistics n	Mean	SD	CV (%)	Min	Median	Max	Geo Mean
Test formulation	t_{max} (h)	30	1.87	1.39	74.4	0.50	1.00	4.00	1.41
	C_{max} (ng/mL)	30	111.5	16.4	14.8	85.07	113.1	155.7	110.3
	AUC_{0-t} (ng·h/mL)	30	3660.89	1352.88	37.0	1060.77	3612.00	6394.69	3369.41
	$AUC_{0-\infty}$ (ng·h/mL)	30	4538.45	1935.68	42.7	1216.51	4286.12	8693.09	4090.34
	$t_{1/2}$ (h)	30	61.28	28.57	46.6	13.64	59.12	123.62	54.25
	MRT (h)	30	83.58	39.08	46.8	16.95	77.45	165.58	73.35
	k_{el} (1/h)	30	0.0149	0.0101	67.5	0.0056	0.0118	0.0508	0.0128
Reference formulation	t_{max} (h)	30	2.13	1.25	58.4	0.50	2.00	4.00	1.78
	C_{max} (ng/mL)	30	105.2	21.3	20.3	79.99	99.92	161.3	103.3
	AUC_{0-t} (ng·h/mL)	30	3729.59	1422.32	38.1	1282.55	3928.45	6314.73	3420.53
	$AUC_{0-\infty}$ (ng·h/mL)	30	4532.40	1863.64	41.1	1538.18	4595.55	8687.36	4117.24
	$t_{1/2}$ (h)	30	58.52	19.99	34.2	15.39	61.89	92.37	54.36
	MRT (h)	30	80.90	30.49	37.7	20.30	88.46	132.09	73.84
	k_{el} (1/h)	30	0.0141	0.0078	55.5	0.0075	0.0112	0.0450	0.0127
Ratio	C_{max} (−)	30	1.097	0.249	22.7	0.689	1.081	1.515	1.068
	AUC_{0-t} (−)	30	1.004	0.203	20.3	0.658	0.982	1.497	0.985

Formulation: Test formulation, 10mg tablet x 2; reference formulation 20mg tablet x 1
N: Number of subjects used to calculate descriptive statistics of each parameter.
Ratio: Test formulation / reference formulation
Geo Mean: Geometric mean

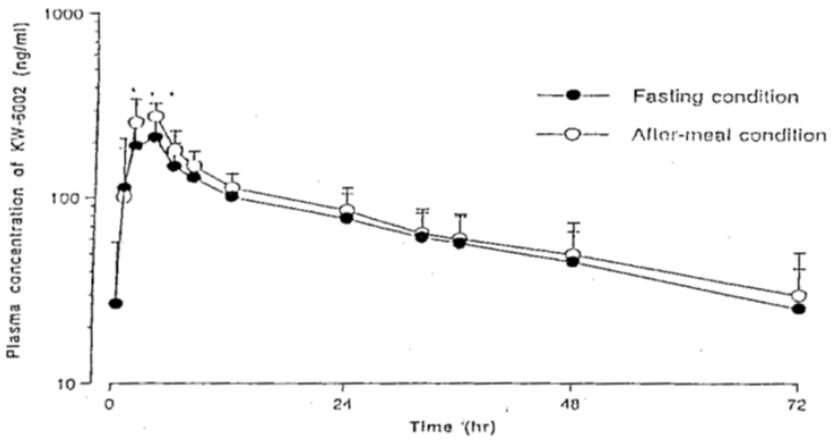
- **Influence of food**

Study 6002-9601 was a placebo-controlled, SB cross-over study with a two-week washout to investigate the effect of feeding on istradefylline PK following a single oral 50 mg dose in healthy young adult male Japanese volunteers.

The plasma concentration-time profiles following istradefylline (KW-6002) administration in fed versus fasted state are presented in the figure below. C_{max} was significantly increased in the fed versus fasted state by approximately 1.4 fold (313.7 ± 42.3 versus 225.5 ± 55.2 ng/mL). While the difference in

$AUC_{0-\infty}$ between fed and fasted participants was not statistically significant (7848 ± 4200 and 6122 ± 2696 ng·hr/ml), feeding increased istradefylline $AUC_{0-\infty}$ by approximately 1.3 fold.

Figure 13: Plasma concentration-time profiles of KW-6002 after a single oral administration of KW-6002 at a dose of 50mg to healthy volunteers



Each point with a bar represents mean + SD of 12 volunteers
* Significantly different from the value of fasting condition ($p<0.05$, paired t-test)
** Significantly different from the value of fasting condition ($p<0.01$, paired t-test)

Study 6002-US-023 was a single-center, OL, randomized, 2-period, 2-sequence, crossover study to investigate the PK parameters of istradefylline following administration of a 40-mg dose under fed (standardised high-fat meal) and fasted conditions in non-smoking, healthy, male subjects.

C_{max}, $AUC_{0-\infty}$ and AUC_{0-t} were all significantly increased in fed versus fasted conditions. The 90% CIs of the fed: fasted ratios for the least squares mean $AUC_{0-\infty}$ and C_{max} were 114.15% - 137.55% and 149.49% - 179.64%, respectively (

Table 6).

Table 6: Analysis of LSM PK parameters for Istradefylline

PK Parameter	Fed	Fasted	Ratio (A/B)	90% CI	P-value
$AUC_{0-\infty}$ [a]	7609.51	6072.63	1.2531	114.15, 137.55	0.0004
AUC_{0-t} [b]	7040.38	5593.00	1.2588	112.95, 140.29	0.0014
C_{max} [b]	263.77	160.96	1.6387	149.49, 179.64	< 0.0001
$t_{1/2}$ [a]	78.2413	68.4524	1.1430	99.66, 128.94	0.1076
$AUC_{0-\infty}$ [c]	7804.63	6218.96	1.2550	114.28, 137.81	0.0004
$t_{1/2}$ [c]	81.6088	71.9278	1.1346	99.51, 127.41	0.1118

a N=24 in fed condition
b Parameter is summarised for the 25 subjects who completed the study under both fed and fasted conditions
c Parameter is summarised for the 23 subjects who completed the study under both fed and fasted conditions and who had estimate $t_{1/2}$ and $AUC_{0-\infty}$ values under both conditions
Note AUCs and C_{max} were back-transformed to original scale from log-transformed values.

Study 6002-011 was an OL, randomized, 2-period, 2-sequence, crossover study to investigate PK of the final istradefylline formulation following administration of a single 20-mg dose under fed and fasted conditions in non-smoking, healthy Japanese adult males.

The C_{max} and $AUC_{0\ t}$ values were approximately 10 to 20% higher under fed versus fasting conditions

Table 7: Summary of plasma PK parameters

Administration Condition	Pharmacokinetic Parameter	Descriptive Statistics							
		n	Mean	SD	Coefficient of Variation (%)	Minimum	Median	Maximum	Geometric Mean
Fed	t_{max} (h)	20	3.10	2.04	65.9	0.50	3.00	8.00	2.39
	C_{max} (ng/mL)	20	136.4	36.0	26.4	82.25	135.2	201.4	131.9
	AUC_{0-t} (ng·h/mL)	20	3833.03	1465.22	38.2	1371.31	3956.63	6082.01	3528.87
	$AUC_{0-\infty}$ (ng·h/mL)	20	4590.54	1997.41	43.5	1698.79	4362.20	8475.15	4159.67
	$t_{1/2}$ (h)	20	53.56	22.33	41.7	24.18	48.94	109.18	49.51
	MRT (h)	20	72.98	32.21	44.1	30.05	66.93	154.00	66.66
	k_{el} (1/h)	20	0.0151	0.0062	40.9	0.0063	0.0142	0.0287	0.0140
Fasting	t_{max} (h)	20	2.23	1.28	57.6	0.50	2.00	4.00	1.87
	C_{max} (ng/mL)	20	112.9	24.1	21.3	77.17	103.5	154.5	110.5
	AUC_{0-t} (ng·h/mL)	20	3397.45	1373.02	40.4	1301.98	3311.94	5826.03	3106.84
	$AUC_{0-\infty}$ (ng·h/mL)	19	4323.23	1990.64	46.0	1680.55	3801.52	9583.48	3912.87
	$t_{1/2}$ (h)	19	57.09	31.51	55.2	21.28	49.03	151.51	50.55
	MRT (h)	19	77.59	43.62	56.2	24.87	66.58	213.01	68.29
	k_{el} (1/h)	19	0.0154	0.0075	49.0	0.0046	0.0141	0.0326	0.0137
Ratio	C_{max} (–)	20	1.219	0.265	21.7	0.823	1.149	1.920	1.193
	AUC_{0-t} (–)	20	1.155	0.209	18.1	0.680	1.164	1.467	1.136

N: Number of subjects used to calculate descriptive statistics of each parameter.
Ratio: Fed-to-fasting ratio

Distribution

The volume of distribution (Vd) of istradefylline at steady state ranged from 448 to 670 L and was independent of the dose administered. The extensive Vd of istradefylline is consistent with its high lipophilicity.

Istradefylline exhibited a high degree of plasma protein binding (mean of approximately 98%), which was independent of the istradefylline concentration in plasma across the anticipated therapeutic concentration range. The dominant binding protein was serum albumin (95%).

Elimination

- **Excretion**

In the mass balance study (6002-US-010), the predominant radiolabelled component in plasma was istradefylline, accounting for >50% of the plasma radioactivity. The overall mean recovery of administered radioactivity (0-432 h interval; [18 days]) was 86.89 ± 8.61%. A total of 38.90 ± 10.70% and 47.99 ± 13.36% of the administered radioactivity was recovered in urine and faeces, respectively. Over the interval 0-168 h (7 days), the corresponding values for urine and faeces were 34.15 ± 10.12% and 39.55 ± 14.33%, respectively.

In urine, unchanged [^{14}C]-istradefylline was not detected. In faecal extracts, unchanged [^{14}C]-istradefylline was the predominant radiolabelled component, accounting for 6.53-65.03% of the faecal radioactivity or 1.25-31.55% of the administered dose.

- **Metabolism**

In vitro studies indicated that the main CYP isoenzymes responsible for the metabolism of istradefylline were CYP1A1 and 3A4/5 isoforms, with CYP1A2, 2B6, 2C8, 2C9, 2C18 and 2D6*1 involved to a much smaller extent (97-015; 97-016; B-168'955; d-06-104; d-07-038). The primary oxidative metabolites of istradefylline were M1 (pharmacologically active, with similar binding affinity for A_{2A} receptors as istradefylline) and M8.

In the mass balance study (6002-US-010), six metabolites, M1, M4, M5, M8, M11, and M12 were observed in plasma extracts. Seven metabolites, M4, M5, M11, M12, M15 (1-β-hydroxylated-3', 4'-O-didemethyl-hydrogenated istradefylline monosulfate), M16 (1-deethyl-4'-O-demethyl istradefylline sulfate), and M19 (3', 4'-O-didemethyl-hydrogenated istradefylline monosulfate) were identified in urine. 5 metabolites M10, M13 (1-deethyl-3', 4'-O-didemethyl-hydrogenated istradefylline), M14 (1-β-hydroxylated-3', 4'-O-didemethyl-hydrogenated istradefylline), M17 (1-β-carboxylated istradefylline), and M18 (3', 4'-O-didemethyl-1-β-carboxylated-hydrogenated istradefylline) were identified in the faeces. Overall, none of the metabolites accounted for >10% of parent drug exposure.

The major routes of istradefylline metabolism identified in humans include: i) reduction of the aliphatic double bond; ii) 3' and/ or 4' -O-demethylation, followed by sulphatation or glucuronidation; iii) oxidation on 1-ethyl to form hydroxylated or carboxylated derivatives; and iv) N-dealkylation to remove 1-ethyl moiety.

- **PK of metabolites**

Quantification of the AUC of M1 and M8 during a dosing interval at steady-state (healthy, non-smoking subjects in Study 6002-US-016) indicated that none of the metabolites could be considered major; the AUCs for the M1 and M8 metabolites were less than 3% and 6%, respectively, of the AUC for the parent drug following repeated dosing of 40 mg/day for 14 days (Table 8). Although M1 is an active metabolite, its concentrations in plasma were very low, thus, no significant contribution to the pharmacological activity of istradefylline is expected.

Table 8: Steady-state PK parameters of M1 (left) and M8 (right) in healthy non-smoking subjects following repeated dosing of 40 mg/day (Study 6002-US-016)

Parameter	Statistic	Healthy Nonsmoker
	n	7
AUC_{0-24} (ng·h/mL)	Mean	120.58
	n	3
$AUC_{0-\infty}$ (ng·h/mL)	Mean	1148.23
	n	7
AUC_{0-t} (ng·h/mL)	Mean	1174.28
	n	7
C_{max} (ng/mL)	Mean	6.35
	n	3
$t_{1/2}$ (hour)	Mean	142.89

Parameter	Statistic	Healthy Nonsmoker
	n	7
AUC_{0-24} (ng·h/mL)	Mean	566.47
	n	7
$AUC_{0-\infty}$ (ng·h/mL)	Mean	4575.41
	n	7
AUC_{0-t} (ng·h/mL)	Mean	4103.47
	n	7
C_{max} (ng/mL)	Mean	31.61
	n	7
$t_{1/2}$ (hour)	Mean	96.74

Dose proportionality and time dependencies

Based on the PK data from studies 6002-US-002, 6002-0104, 6002-9703, PP15710, 6002-US-003, and 6002-US-024, istradefylline exhibited dose proportionality at once-daily doses of up to 80 mg.

Based on the PK data from studies (6002-9703, 6002-0104, 6002-US-002 and 6002-US-003), steady-state is achieved by Day 14 following repeated once daily dosing of istradefylline. Accumulation ratios

(Day 14/Day 1) for AUC0-24 ranged from 2.62 to 5.35 over the istradefylline dose range of 20 to 160 mg/day.

Intra- and inter-individual variability

In Study 6002-US-022, the intra-subject coefficient of variations (CVs) for of $AUC_{0-\infty}$ and C_{max} were 14.1 and 20.7%, respectively, following a single dose of 40 mg istradefylline in non-smoking, healthy adult males.

In the pop-PK analyses [6002-014-pop-pk-r-en (2017) and 6002-nda-response-pop-pk-r-en (2018)], inter-individual variability of istradefylline PK was moderate to high in patients with PD, ranging from 41-43% for CL/F and 40-62% for V1/F.

Pharmacokinetics in the target population

Study 6002-US-003 was a single-center, OL, sequential group study to evaluate the safety and PK of multiple ascending doses of istradefylline administered to PD inpatient volunteers (subjects) under active treatment with L/C. Two doses of istradefylline were administered to separate groups of subjects: 60 and 80 mg/day. Each dose group contained 5 subjects, all of whom received a once daily dose of istradefylline for 14 days.

Table 9: Summary of KW-6002 PK parameters on days 1 and 14

		60 mg dose group (n=5)		80 mg dose group (n=5)	
Parameter	**Units**	**Day 1**	**Day 14**	**Day 1**	**Day 14**
$AUC_{(0-24)}$	ng*h/mL	3085.82 (1082.57)	11475.19 (3463.80)	4365.83 (1336.83)	14971.16 (5304.75)
C_{24}	ng/mL	77.08 (23.33)	430.05 (172.07)	145.40 (46.00)	557.83 (250.69)
C_{max}	ng/mL	267.37 (114.20)	702.06 (223.86)	340.99 (102.48)	940.80 (247.65)
T_{max}	h	2.00 (2.00-4.00)	4.00 (2.00-4.00)	2.00 (1.00-4.00)	2.00 (2.00-48.00)
$t_{1/2}$	h	----	68.94 (24.81)	----	64.13 (19.21)
RAUC		----	3.90 (1.32)	----	3.47 (0.70)
RC_{24}		----	5.62 (1.87)	----	3.73 (0.77)
RC_{max}		----	2.78 (0.88)	----	2.81 (0.39)

Data are presented as mean (SD) with the exception of t_{max} presented as median (minimum-maximum)

The PK of istradefylline in patients with PD under treatment with L/C (present study) were comparable to those in healthy volunteers treated with istradefylline alone (Study 6002-US-002). In Study 6002-US-002, the mean AUC_{0-24} values for normal male subjects were 12,018 and 16,536 ng*h/mL following multiple doses of istradefylline at 60 and 80 mg/day for 14 days, respectively. The average exposure (AUC_{0-24}) of istradefylline in PD patients in the present study was 11,475 and 14,971 ng*h/mL under the same dosing regimens. The mean istradefylline C_{max} after 14 days of multiple dosing were also similar in these two studies: 720 ng/mL in normal male subjects versus 702 ng/mL in PD patients at istradefylline doses of 60 mg/day; 956 ng/mL in normal male subjects versus 941 ng/mL in PD patients at istradefylline doses of 80 mg/day. These results suggest that disposition of istradefylline does not

vary between PD patients and normal male volunteers and that L/C at doses of 500-1500 mg/day does not alter the PK of istradefylline.

Summary statistics for of levodopa and carbidopa PK parameters are provided in Table 10 and Table 11, respectively. There was a decreasing trend in mean levodopa and carbidopa exposure upon co-administration with istradefylline, which became more obvious when multiple doses of istradefylline were given for 14 days.

Table 10: Summary of levodopa PK parameters

		KW-6002 60 mg dose group (n=5)			KW-6002 80 mg dose group (n=5)		
Parameter	**Units**	**Day -1**	**Day 1**	**Day 14**	**Day -1**	**Day 1**	**Day 14**
AUC_{0-2}	ng*h/mL	2489 (2249)	1563 (985)	1209 (915)	2688 (792)	2116 (1354)	1950 (1642)
AUC_{0-4}	ng*h/mL	2842* (1725)	2531* (492)	1983* (1151)	4589**	4396**	2469**
AUC_{1-4}	ng*h/mL	2137* (1331)	2067* (390)	1669* (985)	2520**	3895**	2310**
C_{max}	ng/mL	1595 (1501)	1247 (502)	972 (574)	1845 (421)	1773 (806)	2141 (1207)
T_{max}	h	1 (1-1)	2 (1-4)	1 (1-4)	0 (0-2)	2 (1-4)	2 (1-4)

Data are presented as mean (SD) with the exception of t_{max} presented as median (minimum-maximum)
*n=4
**n=1

Table 11: Summary of Carbidopa PK parameters

		KW-6002 60 mg dose group (n=5)			KW-6002 80 mg dose group (n=5)		
Parameter	**Units**	**Day -1**	**Day 1**	**Day 14**	**Day -1**	**Day 1**	**Day 14**
AUC_{0-2}	ng*h/mL	319 (367)	129 (115)	112* (152)	654 (386)	286 (101)	332 (268)
AUC_{0-4}	ng*h/mL	258* (167)	195* (93)	129** (95)	1474***	948***	416***
AUC_{1-4}	ng*h/mL	171* (105)	158* (70)	113** (74)	855***	824***	311***
C_{max}	ng/mL	196 (205)	108 (63)	79 (77)	440 (154)	217 (98)	238 (154)
T_{max}	h	1 (0-4)	2 (0-4)	3 (1-4)	0 (0-2)	2 (0-2)	2 (1-4)

Data are presented as mean (SD) with the exception of t_{max} presented as median (minimum-maximum)
*n=4
**n=3
***n=1

Special populations

- **Impaired renal function**

Study 6002-US-015 was a multicenter, OL, parallel-group study to assess the effect of severe renal impairment on the single-dose PK of istradefylline. After a single oral dose of 40 mg of istradefylline, the ratio of the LSM $AUC^{0-\infty}$ of istradefylline was 84.36% (90% CI: 49.48% to 143.81%) in renally impaired subjects compared with matched healthy subjects, and 89.74% (90% CI: 52.64% to 152.98%) in renally impaired subjects compared with young healthy subjects. Elimination half-life of istradefylline was comparable across the 3 groups of subjects (94.84 to 117.24 hours). No apparent trends in the extent of protein binding were observed.

- **Impaired hepatic function**

Study 6002-016 was a two center, OL, parallel group study to investigate the effect of mild hepatic impairment on the PK of istradefylline. 10 subjects with mild hepatic impairment (Child-Pugh Class A) and 10 subjects with normal hepatic function received a single dose of istradefylline 40 mg. The geometric mean ratio of C_{max} and $AUC_{0-\infty}$ were 70.2% (90% CI: 51.7% to 95.2%) and 90.9% (90% CI: 56.7% to 145.8%) in the subjects with mild hepatic impairment compared to the subjects with normal hepatic function. The effect of mild hepatic impairment on the exposure of metabolites was variable; no effect on istradefylline M1 metabolite and 27% lower exposure to istradefylline M8 metabolite. Given the low abundance of metabolites in the plasma (<10%), these differences are not of clinical relevance.

Study 6002-US-016 was a phase 1, multicentre, OL, parallel-group study to evaluate the influence of moderate hepatic impairment (Child-Pugh Class B) on the PK of istradefylline. Twenty-eight subjects were enrolled in 4 groups (7 subjects per group): hepatically impaired smokers (at least 20 cigarettes per day); healthy smokers (at least 20 cigarettes per day); hepatically impaired nonsmokers; healthy nonsmokers. All patients received an oral dose in tablet form of 40 mg istradefylline once daily for 14 days.

The results of the statistical analyses are provided in Table 12 and Table 13. The LSM AUC0-∞ ratio was 1.50 (90% CI: 59.53 to 376.63) in Hepatically Impaired Smokers/Healthy Smokers, and 1.14 (90% CI: 42.17 to 309.30) in Hepatically Impaired Nonsmokers/Healthy Nonsmokers.

Table 12: Statistical evaluation of the difference in Istradefylline PK parameters between hepatically impaired smokers and healthy smokers

Parameter	Statistic	Hepatically Impaired Smoker	Healthy Smoker	Ratio[a,c]	90% CI[b]
AUC_{0-24} (ng·h/mL)	n	7	7		
	Mean	5183.50	5205.64	1.00	64.25, 154.32
$AUC_{0-\infty}$ (ng·h/mL)	n	7	7		
	Mean	22432.15	14981.03	1.50	59.53, 376.63
AUC_{0-t} (ng·h/mL)	n	7	7		
	Mean	21077.11	14682.34	1.44	62.91, 327.58
C_{max} (ng/mL)	n	7	7		
	Mean	351.68	379.72	0.93	62.66, 136.88
$t_{1/2}$ (hour)	n	7	7		
	Mean	99.63	55.55	1.79	1.52, 357.14

Note: For AUCs and C_{max} mean was evaluated by exponentiating the LSM of the $\log_e$ transformed PK parameter. For $t_{1/2}$ mean was the LSM on original scale. The LSM is from the ANOVA model
a For AUCs and C_{max}, ratio and CIs are based on back log-transformation of the log-transformed data. For $t_{1/2}$, ratio and Cis are based on original data. To obtain percentage multiply value by 100.
b 90% CI for mean group differences was calculated from ANOVA. Limits of the Cis expressed as percentages
c To obtain percentage multiply value by 100.

Table 13: Statistical evaluation of the difference in Istradefylline PK parameters between hepatically impaired nonsmokers and healthy nonsmokers

Parameter	Statistic	Hepatically Impaired Nonsmoker	Healthy Nonsmoker	Ratio[a,c]	90% CI[b]
	n	7	7		
AUC_{0-24} (ng·h/mL)	Mean	8111.52	8913.93	0.91	58.72, 141.03
	n	6	6		
$AUC_{0-\infty}$ (ng·h/mL)	Mean	96802.41	84758.02	1.14	42.17, 309.30
	n	7	7		
AUC_{0-t} (ng·h/mL)	Mean	72607.91	71088.61	1.02	44.76, 233.07
	n	7	7		
C_{max} (ng/mL)	Mean	489.26	479.11	1.02	69.09, 150.93
	n	6	6		
$t_{1/2}$ (hour)	Mean	287.61	117.99	2.44	153.33, 334.18

Note: For AUCs and C_{max} mean was evaluated by exponentiating the LSM of the loge transformed PK parameter. For $t_{1/2}$ mean was the LSM on original scale. The LSM is from the ANOVA model
a For AUCs and C_{max}, ratio and CIs are based on back log-transformation of the log-transformed data. For $t_{1/2}$, ratio and Cis are based on original data. To obtain percentage multiply value by 100.
b 90% CI for mean group differences was calculated from ANOVA. Limits of the Cis expressed as percentages
c To obtain percentage multiply value by 100.

The fraction at steady state (fss) value was 97% in the healthy smokers, 89% in the hepatically impaired smokers and 86% in healthy nonsmokers, indicating that plasma concentrations were at or near steady-state by Day 14. In contrast, steady-state for istradefylline in hepatically impaired nonsmokers was not attained by Day 14 as indicated by the fss value of 62%. Therefore, steady-state PK values for the hepatically impaired non-smoker group were estimated to support inter-group comparisons.

Initially, steady-state exposure in hepatically impaired nonsmokers was estimated using the accumulation ratio calculated from the $t_{1/2}$ of istradefylline and the estimated average drug concentration at steady-state. According to this analysis, Area Under the plasma Concentration versus time curve from 0 to 24 hours (AUC_{0-24h}) at steady state in hepatically impaired nonsmokers was estimated to be around 3.3-fold higher than in healthy nonsmokers. However, the use of $t_{1/2}$ of the concentration-time profile to predict accumulation at steady-state, which may not be representative of the $t_{1/2}$ of istradefylline, may have resulted in an over-estimation of the steady-state exposure.

To provide a more accurate estimate of the effect of moderate hepatic impairment on istradefylline PK, re-analysis based on compartmental PK modelling and simulation was conducted. Based on this analysis, smoking subjects with moderate hepatic impairment and smoking healthy subjects showed comparable drug exposure at steady-state as indicated by the ratio (1.03) of the mean AUC_{0-24h} values on Day 120. In nonsmokers, there was a 39% increase in AUC_{0-24h} on Day 120 in hepatically impaired subjects compared to healthy subjects (Table 14). This increase in steady-state exposure in subjects with hepatic impairment was considered unlikely to be of clinically relevance.

Table 14: Re-analysis: the effect of moderate hepatic impairment and smoking on steady-state exposure ($AUC_{0\text{-}24h}$) to Istradefylline based on 2-Compartment PK modelling and simulation (Study 6002-US-016)

Comparison	Mean Ratio of $AUC_{0\text{-}24hr}$ on Day 120	90%Confidence Interval (CI)
Effect of Moderate Hepatic Impairment (Ratio of Subjects with Moderate Hepatic Impairment *versus* Healthy Subjects)		
Smokers	1.03	(0.49, 2.19)
Nonsmokers	1.39	(0.93, 2.19)
Effect of Cigarette Smoking (Ratio of Smokers *versus* Nonsmokers)		
Subjects with Moderate Hepatic Impairment	0.462	(0.20, 0.78)
Healthy Subjects	0.622	(0.33, 1.01)

- **Gender**

Study 6002-EU03 was a single-centre, randomised, DB, placebo-controlled, 3-way cross-over study to investigate the PK of istradefylline following three single ascending doses of 50, 100 and 150 mg in 19 healthy male and female elderly volunteers aged (n=9 males and 10 females).

Istradefylline (KW-6002) exposure was higher in females, with AUC values greater at all doses administered (Table 15). The $t_{1/2}$ of istradefylline in female subjects was approximately 2-fold higher than males. Statistical examination of the terminal rate constants indicated that there was a significant sex difference, with λz significantly lower in females (p = 0.046).

Table 15: Mean (SD) PK parameters of KW-6002 following administration of single oral doses to male and female elderly subjects

Parameter	KW-6002 dose administered					
	50 mg		100 mg		150 mg	
	Males	Females	Males	Females	Males	Females
C_{max} (ng/ml)	171.4 (28.4)	195.7 (38.7)	293.0 (48.8)	261.4 (106.6)	302.6 (79.5)	341.0 (112.4)
T_{max} (hours)[a]	2.0	2.5	3.5	2.5	2.5	3.0
AUC_{all} (ng·h/ml)	7534 (5293)	12575 (6695)	15506 (11051)	21192 (8147)	21681 (13991)	35665 (13583)
AUC (ng·h/ml)	7853 (5954)	14592 (7604)	15888 (11803)	21509 (8465)	21960 (14694)	36699 (15303)
AUC_t (ng·h/ml)	7472 (5323)	12560 (6713)	15437 (11105)	21182 (8165)	21444 (14154)	35636 (13591)
λ_z ($hours^{-1}$)	0.0174 (0.0100)	0.0075 (0.0030)	0.0195 (0.0106)	0.0101 (0.0029)	0.0161 (0.0103)	0.0077 (0.0032)
$t_{½}$ (hours)[b]	39.9	92.4	35.5	68.6	43.1	90.0

a Value quoted is the median
b Calculated as ln2/mean λz

Statistical analysis indicated that there was a significant effect of smoking on C_{max} (p = 0.012), AUC (p = 0.007) and terminal rate constant (p = 0.019) in male subjects with a lower C_{max} and AUC and a higher terminal rate constant observed in the male smokers. There was no significant sex effect in non-smokers for C_{max} (p = 0.45), AUC (p = 0.87) and terminal rate constant (p = 0.98)

Study PP15710 was a DB, placebo-controlled, randomised, multiple ascending dose study of istradefylline in elderly male and female subjects (age >55 years). The study evaluated a possible gender effect on istradefylline PK (a secondary objective). The study population was divided into 4 groups: Groups 1 to 3 (male and female non-smokers) and Group 4 (male smokers). Istradefylline was administered once daily for 14 consecutive days: 10 mg/day (Group 1), 20 mg/day (Group 2), 40 mg/day (Groups 3 and 4), or placebo. In Groups 1 to 3, 12 subjects (6 males and 6 females) were randomised to istradefylline. The rate and extent of systemic exposure of female subjects to istradefylline were generally similar to those in male subjects (Table 16) and there was no statistically significant evidence for any sex-related differences in systemic exposure (p≥0.843).

Table 16: Mean C_{max} and AUC_{24} Ratios (Males to Females) and Ro 64-4529 and their 95% CI. Summary of Groups 1, 2 and 3.

Parameter	Ratio	Lower	Upper
C_{max}	0.98	0.81	1.18
AUC_{24}	1.02	0.82	1.28

In the TQT Study 6002-US-024, the effect of sex on the PK of istradefylline was evaluated following 14-day oral administration of istradefylline at doses of 40 and 160 mg to 22 males and 22 females in each treatment group. In the analysis of variance (ANOVA) model, the gender-by-treatment interaction for istradefylline was not significant following single dose and multiple dose administration.

Using the final pop-PK models [6002-014-pop-pk-r-en (2017) and 6002-nda-response-pop-pk-r-en (2018)], simulations were conducted to evaluate the impact of sex on istradefylline steady state exposure ($AUC_{0-\tau,ss}$) at a dose of 20 mg/day. Female sex was associated with a 14-32% increase in istradefylline exposure.

- **Race**

Using the final pop-PK model [6002-nda-response-pop-pk-r-en (2018)], simulations were conducted to evaluate the impact of Asian race on istradefylline steady state exposure ($AUC_{0-\tau,ss}$) at a dose of 20 mg/day. Non-Asian race was associated with a 35% decrease in istradefylline exposure relative to the reference Asian subject.

- **Weight**

Using the final pop-PK model [6002-014-pop-pk-r-en (2017)], simulations were conducted to evaluate the impact of the BMI on istradefylline steady state exposure ($AUC_{0-\tau,ss}$) at a dose of 20 mg/day. Predicted exposure was 15% lower and 21% higher in males with low and high BMI, respectively, compared to a typical male subject with a BMI of 26.6 kg/m2 (Table 17).

Table 17: Impact of covariates on istradefylline exposure

CL/F (mL/hr)	AUC(0-τ)ss (ng·hr/mL)	Description[a]	BMI (kg/m^2)	Sex	AUC(0-τ)ss %Reference
4.82	4536.9	Reference	26.6	Male	100%
4.21	5192.8	Female	26.6	Female	114%
5.68	3851.5	Low BMI	20.2	Male	85%
3.99	5476.9	High BMI	36.5	Male	121%
4.96	4408.3	Low BMI	20.2	Female	97%
3.49	6268.5	High BMI	36.5	Female	138%

a Low and high covariate values are the 5th and 95th percentiles of their respective baseline distributions. The reference subject is a male with median BMI

Using the final pop-PK model [6002-nda-response-pop-pk-r-en (2018)], simulations were conducted to evaluate the impact of weight on istradefylline steady state exposure ($AUC_{0-\tau,ss}$) at a dose of 20 mg/day. Predicted exposure was 34% higher and 26% lower in male subjects at the high and low ends of the weight distribution, respectively, relative to the 65 kg, male reference subject (Table 18).

Table 18: Impact of covariate on exposure

CL/F (L/hr)	AUC(0-τ)ss (ng·hr/mL)	Description [a]	Weight (kg)	Bilirubin (mg/dL)	Race	Sex	AUC(0-τ)ss %Reference
5.09	4282.7	Reference	65.0	0.53	Asian	Male	100%
3.86	5655.5	Female	65.0	0.53	Asian	Female	132%
7.82	2788.5	Non-Asian	65.0	0.53	Non-Asian	Male	65%
6.88	3172.6	Low Weight	41.0	0.53	Asian	Male	74%
3.80	5742.8	High Weight	102.0	0.53	Asian	Male	134%
5.59	3902.9	Low Bilirubin	65.0	0.29	Asian	Male	91%
4.62	4722.7	High Bilirubin	65.0	1.0	Asian	Male	110%

a Reference subject is an Asian male with median weight and bilirubin. Low and high weight and bilirubin values are the 5th and 95th percentiles of the baseline distribution. All simulated subjects dosed once daily to steady-state with 20mg KW6002.

- **Elderly**

Study 6002-0205 was an OL parallel group study to investigate the effect of age on istradefylline (KW-6002) and metabolite M1 PK. Healthy elderly and non-elderly adult males (n=9 per group) received a single 40 mg oral dose of istradefylline in the fasted state. There were no statistically significant differences in t_{max}, C_{max} and AUC_{0-t} between elderly and non-elderly participants.

Table 19: Summary statistics for KW-6002 plasma PK parameters

Treatment	Summary Statistics	t_{max} (h)	C_{max} (ng/mL)	AUC_{0-t} (ng·h/mL)	$AUC_{0-\infty}$ (ng·h/mL)	Vd/F (L)	CL/F (L/h)	MRT (h)	$t_{1/2}$ (h)	kel (/h)
A	N	9	9	9	4	4	4	4	4	4
(Elderly)	Mean	2.67	170.03	5469.3	-	-	-	-	-	-
	SD	1.00	47.65	2990.8	-	-	-	-	-	-
	Median	2.00	153.20	4153.0	-	-	-	-	-	-
	Minimum	2.0	106.5	2109	4604	253	4.24	57.1	41.4	0.0110
	Maximum	4.0	244.4	10834	9440	762	8.69	76.5	63.2	0.0167
	Geometric Mean	2.52	164.08	4811.1	-	-	-	-	-	-
	CV	37.50	28.02	54.68	-	-	-	-	-	-
B	N	9	9	9	8	8	8	8	8	8
(Young)	Mean	2.78	172.58	4410.6	4869.4	382.6	10.828	43.01	32.39	0.02974
	SD	1.20	30.32	2271.8	2641.0	91.7	5.881	25.67	19.40	0.01708
	Median	2.00	177.30	3634.0	4020.5	394.5	10.650	34.40	25.80	0.02780
	Minimum	1.0	128.7	1743	1901	242	4.65	18.5	12.5	0.0113
	Maximum	4.0	209.8	8093	8606	552	21.05	80.7	61.3	0.0553
	Geometric Mean	2.52	170.11	3904.0	4240.8	372.9	9.434	36.59	27.41	0.02530
	CV	43.27	17.57	51.51	54.24	23.95	54.31	59.67	59.91	57.45

Overall, the Phase 1 clinical pharmacology studies and pop-PK results suggest that istradefylline exposure estimates were relatively similar among the age categories for the elderly subpopulations. It should be noted that in the Phase 1 PK studies there were no subjects aged 85+ (Table 20), and in the population analyses, there were a total of 4 individuals who had exposure estimates in this subgroup: 2 from Study 6002-US-006 and 2 from Study 6002-014.

Table 20: Subjects in phase 1 clinical pharmacology studies by age category

	Age 65-74 (Number of subjects / total number)	Age 75-84 (Number of subjects / total number)	Age 85+ (Number of subjects / total number)
PK trials[a]	46/943	9/943	NA/943
Applicable PK trials[b]	44/189	8/189	NA/189

PK = pharmacokinetic.

a 28 PK trials: Studies 6002-EU01, 6002-9601, 6002-EU03, 6002-0205, 6002-US-022, 6002-012, 6002-US-023, 6002-011, 6002-US-010, 6002-EU02, 6002-9703, 6002-0104, 6002-US-002, EUPP15710, 6002-EU06, 6002-US-024, 6002-US-003, BP15748, 6002-US-009, BP15809, 6002-US-008, 6002-015, 6002-US-020, 6002-US-026, 6002-US-015, 6002-016, 6002-017, 6002-US-016.
b PK trials that include older subjects (6002-EU03, 6002-0205, PP15710, 6002-EU04, 6002-015, 6002-US-003, 6002-US-015, 6002-016, and 6002-US-016)

Pharmacokinetic interaction studies

- ***In vitro***

CYP isoenzymes

Istradefylline exhibited no significant inhibition of the CYP1A2, 2B6, 2C9, 2C19 and 2D6 activities in human microsomes. However, istradefylline inhibited irreversibly the CYP3A4/5 activity (B-168'955; 99-146; d-04-044; d-06-178; 2002-063A).

In sandwich-cultured human hepatocytes, istradefylline at concentrations from 0.1 to 30 µmol/L did not induce CYP1A2 and 2B6 mRNA levels, but caused concentration-dependent increases in CYP3A4 mRNA levels, suggesting that istradefylline is likely an *in vitro* inducer for CYP3A4 (XT-153111).

Transporters

In a Caco-2 cell system, istradefylline was found to be highly permeable and not a substrate for P-gp but an inhibitor of P-gp (d-05-194; d-06-071).

In vitro studies indicated that istradefylline was neither a substrate for BCRP in transporter-expressing LLC-PK1 cells nor a substrate for OATP1B1 and OATP1B3 in transporter-expressing HEK-293 cells (GE-1451-G).

The ratios of gut luminal concentration to inhibition potencies (Igut/IC50) of istradefylline for human P-P-gp and human BCRP were 239 and 6090, respectively, and both values greatly exceeded the cut-off value of 10, suggesting that istradefylline has the potential to inhibit P-gp and BCRP *in vivo* (r-18-0051).

In vitro studies suggested that istradefylline did not have an inhibitory effect on OAT3 but had an inhibitory effect on drug transporters OATP1B1, OATP1B3, OAT1, OCT2, MATE1, and MATE2-K. However, all calculated values regarding DDI risk assessment using basic models were below the cut-off values, suggesting minimal DDI risk of istradefylline for *in vivo* inhibition on the transporters (r-18-0051).

- ***In vivo***

Study BP15809 was an OL, multiple dose study to investigate the effect of istradefylline on the PK of the CYP3A substrate midazolam. 24 healthy, male subjects received a single oral 7.5 mg dose of midazolam on Day 1. After a washout period of 48 hours, on Day 3, treatment with oral istradefylline 5 mg (Treatment Group 1) or 20 mg (Treatment Group 2) once daily for 14 days was started. On the last day of treatment with istradefylline (Day 16), subjects received a single oral 7.5 mg dose of midazolam.

The C_{max} values of midazolam in healthy subjects on Day 16, after 14 days treatment with istradefylline were generally similar to those on Day 1 (in the absence of istradefylline). The 90% CI of the mean C_{max} ratio (Day 16/Day 1) were 0.71 to 1.09 and 0.62 to 0.97 in Treatment Group 1 (istradefylline 5mg/day) and Treatment Group 2 (istradefylline 20 mg/day), respectively. The AUC_{0-24h} values of midazolam in healthy subjects in the presence of istradefylline (Day 16) were similar to those in the absence of istradefylline (Day 1). The 90% CIs of the mean AUC_{0-24h} ratio (Day 16/Day 1) were 0.94 to 1.28 and 0.81 to 1.12 in Treatment Group 1 (istradefylline 5 mg/day) and Treatment Group 2 (istradefylline 20 mg/day), respectively.

Study 6002-US-008 was an OL, sequential, 2-period study to investigate the *in vivo* interaction of istradefylline with CYP3A4 in non-smoking, normal, healthy subjects (male or female) between the ages of 18 and 55 years. The study consisted of 2 parts.

Part 1 (midazolam interaction) was conducted to determine the effect of istradefylline on the PK of midazolam, a CYP3A4 substrate. Subjects received a single oral dose of midazolam 10 mg on Day 1. Starting on Day 4, subjects received daily doses of istradefylline 80 mg for 15 days (Days 4 to 18) to achieve steady state. A single oral dose of midazolam 10 mg was administered again on Day 17.

The mean C_{max} of midazolam was 1.6-fold (90% CI 1.32-1.96) greater and the mean $AUC_{0-\infty}$ was 2.4-fold (90% CI 1.13-2.73) greater for midazolam in the presence of istradefylline as compared with midazolam alone. The $t_{1/2}$ ratio for midazolam in the presence of istradefylline as compared with midazolam alone was 1.08 (Table 21).

Table 21: LSM PK parameters for Midazolam (cohort 1)

Parameter	N	Midazolam (Treatment A)	N	Midazolam + Istradefylline (Treatment B)	Ratio B/A[a]	90% CI
ln C_{max}	16	3.2859	16	3.7639	1.61	1.32 – 1.96
t_{max}[b]	16	1.00[c]	16	1.00[d]	0·00[d]	-0.25 – 1.00[e]
ln $AUC_{0-\infty}$	16	4.8319	16	5.7103	2.41	2.13 – 2.73
ln AUC_{0-48}	16	4.8232	16	5.7030	2.41	2.13 – 2.73
ln AUC_{0-24}	16	4.8012	16	5.6417	2.32	2.05 – 2.62
$t_{1/2}$[f]	16	6.6263	16	7.1827	1.08	0.94 – 1.23
ln CL/F	16	7.1919	16	6.3134	0.42	0.37 – 0.47

a For Log_e transformed parameters $AUC_{0-\infty}$, AUC_{0-48}, AUC_{0-24}, CL/F and C_{max}, ratios of B/A were calculated by equation ratio = $e^{(LSM\ for\ treatment\ B-\ LMS\ for\ treatment\ A)}$. For untransformed parameters, $t_{1/2}$ and t_{max} ratios of B/A were calculated by equation ratio = LSM for treatment B/LSM for treatment A

Part 2 (ketoconazole interaction) was conducted to determine the effect of ketoconazole, a potent CYP 3A4 inhibitor, on the PK of istradefylline. Subjects received a single dose of 40 mg istradefylline on Day 1. On Day 15, subjects were administered oral ketoconazole 200 mg twice daily for 4 days (Day 15 to Day 18). A single dose of istradefylline 40 mg was administered again on Day 19 (Period 2) followed by 200 mg once daily of ketoconazole for 7 days (Day 19 to Day 25).

The ratio (Day 19/Day 1) of the LSM for istradefylline C_{max} was 98.6% (90% CI 85.55-113.66%). There was an approximate 2.5-fold (90% CI 1.92-3.18) increase in AUC0-∞ on Day 19 vs. Day 1 and mean istradefylline half-life increased from 89 hours to about 276 hours. However, since the sampling duration was only for 168 hours, several subjects had extrapolated areas that comprised greater than 60% of the AUC0-∞. Therefore, AUC0-∞ may have been overestimated. AUC0-t and AUC0-24 increased in the presence of ketoconazole, by 1.51-fold (90% CI 1.40-1.63) and 1.15-fold (90% CI 1.07-1.22), respectively (Table 22).

Table 22: LSM PK parameters for Istradefylline (cohort 2)

Parameter	Istradefylline Day 1	Istradefylline Day 19	Ratio Day 19/Day 1	90% CI	p-value
ln C_{max}	288.59	284.57	98.61	85.55 - 113.66	0.8646
t_{max}	3.23	2.60	-0.63	-1.22 - -0.05	0.0777
ln $AUC_{0-\infty}$	12964.5	32004.6	246.86	191.70 - 317.90	< 0.0001[c]
ln AUC_{0-t}	9428.5	14239.1	151.02	140.09 - 162.81	< 0.0001[c]
ln AUC_{0-24}	2970.2	3404.4	114.62	107.47 - 122.25	0.0022[b]
ln AUC_{0-72}	6183.2	8286.3	134.01	128.00 - 140.30	< 0.0001[c]
ln AUC_{all}	9428.5	14239.1	151.02	140.09 - 162.81	< 0.0001[c]
$t_{1/2}$	89.35	276.45	187.09	41.08 - 333.10	0.0405[a]
ln CL/F	51.43	20.85	40.54	31.50 - 52.16	< 0.0001[c]

a Statistically significant at p=0.05
b Statistically significant at p=0.01
c Statistically significant at p=0.001
Note: Analyses on t_{max} and $t_{1/2}$ were performed on untransformed data.

Study 6002-US-020 was a Phase 1, single center, in-patient, DB, placebo-controlled, randomised study in normal, healthy, non-smoking male subjects (18-55 years of age) to evaluate the effect of istradefylline at steady state on the single-dose PK of atorvastatin and its ortho- and para-hydroxy metabolites. 16 subjects received a single oral dose of atorvastatin 40 mg on Day 1, istradefylline 40 mg/day from Days 5 to 21, and a single oral dose of 40 mg atorvastatin on Day 18.

The least squares mean PK parameters for atorvastatin are shown in Table 23. The peak exposure (C_{max}), total exposure (AUC), and $t_{1/2}$ of atorvastatin were all significantly increased in the presence of istradefylline.

Table 23: Statistical analysis of PK parameters for Atorvastatin

PK Parameter	N	Atorvastatin + Istradefylline (Treatment B)	N	Atorvastatin (Treatment A)	Ratio B/A[a]	90% CI
ln $AUC_{0-\infty}$	16	4.88	16	4.45	1.54	137.32 to 171.88
ln AUC_{0-t}	16	4.82	16	4.37	1.57	140.32 to 175.44
ln C_{max}	16	2.63	16	2.20	1.53	133.78 to 175.02
$t_{1/2}$	16	11.99	16	9.46	1.27	111.30 to 142.08
t_{max}[b]	16	2.0	16	2.5	-0.99	-1.5 to -0.5

a For Log_e transformed parameters $AUC_{0-\infty}$, AUC_{0-t}, and C_{max}, ratios of B/A were calculated by equation ratio = $e^{(LSM\ for\ treatment\ B-\ LMS\ for\ treatment\ A)}$. For untransformed parameter $t_{1/2}$ ratios of B/A were calculated by equation ratio = LSM for treatment B/LSM for treatment A
b Median values for t_{max} are listed. For t_{max} the median difference of B-A was calculated.

The LSM values for $AUC_{0-\infty}$ were slightly higher for metabolites ortho-hydroxy atorvastatin and para-hydroxy atorvastatin (~20% and 6%, respectively) in the presence of istradefylline. The presence of istradefylline did not alter the $t_{1/2}$ of these 2 metabolites.

Study 6002-015 was a single-center, OL, one-sequence, two-period study to investigate the effect of multiple doses of rifampicin, (a strong CYP3A4 inducer) on the single-dose PK parameters of istradefylline in 20 healthy non-smoking, adult subjects.

A substantial decrease in exposure to istradefylline (C_{max}, AUC_{0-last}, $AUC_{0-\infty}$) and a reduction in the mean $t_{1/2}$ was observed with coadministration of rifampin and istradefylline as compared to administration of istradefylline alone. The geometric mean ratio of C_{max} and $AUC_{0-\infty}$ were 55.50% (90% CI: 49.45% to 62.29%) and 19.16% (90% CI: 17.87% to 20.54%), respectively, with coadministration of rifampin and istradefylline as compared to administration of istradefylline alone (Table 24).

Table 24: Statistical comparisons of plasma Istradefylline PK parameters

Parameter	Geometric Mean		Geometric Mean Ratio [c]	90% Confidence Interval
	Istradefylline Alone [a]	Istradefylline+Rifampin [b]		
C_{max}	170.8	94.8	55.50	49.45-62.29
$AUC_{0\text{-last}}$	9457.3	1964.6	20.77	19.53-22.10
$AUC_{0\text{-}\infty}$	10575.3	2025.9	19.16	17.87-20.54

a A single 40mg oral dose of istradefylline administered alone on Day 1 in Period 1
b Daily oral doses of 600mg rifampicin from Day 1 to 20 with a single 40mg oral dose of istradefylline on Day 8 in Period 2
c Geometric mean ratio of istradefylline + rifampicin versus istradefylline alone.

The effects of rifampin on the istradefylline M1 and M8 metabolites were inconsistent and variable. However, the rifampicin effect on the 2 metabolites is not considered clinically relevant as the M/P ratio for $AUC_{0\text{-last}}$ or $AUC_{0\text{-}\infty}$ for M1 and M8 were less than 10%.

Study 6002-us-026 was a drug interaction study to evaluate the effect of istradefylline as a potential P-gp inhibitor using digoxin as a P-gp substrate. In this study, 0.4 mg of digoxin was administered either alone or co-administered with istradefylline (40 mg/day for 21 days).

The results indicated a mild drug interaction between steady-state 40 mg/day istradefylline and a single dose of digoxin 0.4 mg. C_{max} of digoxin was 33% higher when istradefylline was co-administered. The mean $t_{1/2}$ of digoxin remained unchanged in the presence of istradefylline as compared to that with digoxin alone. The LSM AUCs of digoxin were 19% to 25% higher when istradefylline was coadministered (Table 25).

Table 25: Statistical evaluation of the effects of Istradefylline on the PK of Digoxin

Parameter (unit)	Statistic	Digoxin + Istradefylline N=21	Digoxin Alone N=21	P-value[a]	Ratio[b]	90% Confidence[c] Interval
$AUC_{0\text{-}\infty}$ (pg.hr/mL)	Mean	27695.52	22960.08		1.2062	111.28, 130.76
	Median	28380.13	24157.19			
	Min, Max	17681.02, 47016.32	12910.16, 38249.09			
$AUC_{0\text{-}48}$ (pg.hr/mL)	Mean	16431.10	13853.82		1.1860	108.13, 130.09
	Median	16726.22	14489.04			
	Min, Max	12402.72, 22229.97	7351.80, 23184.17			
$AUC_{0\text{-}t}$ (pg.hr/mL)	Mean	23772.19	18971.42		1.2531	114.20, 137.49
	Median	25133.24	19916.51			
	Min, Max	15716.27, 38622.36	9038.76, 34515.84			
CL_R (L/hr)	Mean	9.35	11.16		0.8384	76.09, 91.59
	Median	8.48	10.31			
	Min, Max	6.90, 14.14	7.43, 19.31			
C_{max} (pg/mL)	Mean	3386.19	2544.27		1.3309	116.31, 152.29
	Median	3355.13	2458.08			
	Min, Max	2459.60, 5634.24	1115.57, 4304.52			
$t_{1/2}$ (hr)	Mean	42.60	41.77		1.0198	90.33, 113.64
	Median	41.54	41.50			
	Min, Max	23.02, 63.85	23.06, 64.17			
T_{max} (hr)	Mean	0.72	0.74			
	Median	0.52	0.50	0.822		
	Min, Max	0.50, 1.00	0.50, 1.50			

Summary statistics, CIs and p-value are based on log-transformed data for AUCs and C_{max} and original data for CL_R and $t_{1/2}$.
AUCs and C_{max} means were calculated by exponentiating the LSM of the $\log_e$ transformed PK parameters
a From a Wilcoxon Rank Sum test of medians
b Mean ratio for Digoxin + Istradefylline/Digoxin alone (percentage = ratio x 100).
c 90% CI for mean group differences was calculated from ANOVA. CIs are expressed as percentages.

Study BP15748

Study BP15748 was a DB, placebo-controlled, randomised DDI study between repeated doses of L/C and repeated doses of istradefylline in 32 healthy, non-smoking, male and female subjects aged 18 to 40

years. Subjects were treated openly with multiple doses of L/C (100/25 mg) for three weeks (Days 1-21, t.i.d). On Day 8, treatment with istradefylline (20 or 40 mg/day) or placebo began in a double-blind, randomised fashion for 14 days (Days 8-21).

The C_{max0-6}, $C_{max6-12}$, AUC_{0-6}, AUC_{6-12} and AUC_{0-12} values of levodopa on Day 21, after 14 days treatment with istradefylline, were generally similar to those indices of exposure on Day 7 (in the absence of istradefylline). The mean ratios for AUC_{0-12} (Day 21 to Day 7) did not differ significantly from 1 with either the 20 mg istradefylline dose (1.07 [95% CI: 0.97, 1.17]) or the 40 mg istradefylline dose (0.93 [95% CI: 0.85, 1.02]). On Day 21, the PK values for levodopa in the placebo group were generally similar to the values in the istradefylline group.

The C_{max0-6}, $C_{max6-12}$, AUC_{0-6}, AUC_{6-12} and AUC_{0-12} values of carbidopa on Day 21, after 14 days treatment with istradefylline, were generally similar to those indices of exposure on Day 7 (in the absence of istradefylline). The mean AUC^{0-12} ratio (Day 21/Day 7) did not differ significantly from 1 in subjects receiving either 20 or 40 mg istradefylline.

A comparison of Day 14 C_{max} and AUC_{0-24} of istradefylline obtained from study PP15710 (a multiple ascending dose study in elderly subjects) with those from the current study BP15748 suggested that istradefylline 40 mg/day combined with L/C had a 15% lower C_{max} and an 11% lower AUC compared to istradefylline treatment alone, which was not statistically significant.

Study 6002-US-009 was a single-center, OL, sequential drug interaction study to evaluate the effects of 14 days of oral administration of 80 mg istradefylline on the PK of a single dose of L/C in normal, non-smoking, healthy subjects. 24 subjects received single oral doses of L/C (200 mg/50 mg) on Days 1 and 15, and daily oral doses of 80 mg istradefylline for 14 days (Day 2 through Day 15). The PK parameters of levodopa and carbidopa are shown in Table 26.

Table 26: PK parameters of Levodopa and Carbidopa following daily doses of 80mg Istradefylline for 14 days (Study 6002-US-009)

Parameter	Without Istradefylline (Day 1) (N = 24)	With Istradefylline (Day 15) (N = 24)	Difference or Ratio[a] (%) (90% CI)
Levodopa			
T_{max} (h)	0.760 (60.1)	0.875 (53.9)	0.115 (−0.107, 0.336) [b]
C_{max} (ng/mL)	1794 (31.4)	2005 (21.0)	111.7 (100.9, 123.7)
AUC_{0-12} (ng·h/mL)	4029 (23.8)	4262 (19.4)	105.8 (101.4, 110.4)
$t_{1/2}$ (h)	1.57 (8.5)	1.52 (8.8)	−0.051(−0.087, −0.016) [b]
Carbidopa			
T_{max} (h)	3.02 (33.9)	2.66 (44.0)	−0.365 (−0.917, 0.188)[b]
C_{max} (ng/mL)	247 (39.9)	267 (40.3)	108.0 (99.1, 117.7)
AUC_{0-12} (ng·h/mL)	1116 (34.3)	1145 (39.5)	102.6 (95.5, 110.3)
$t_{1/2}$ (h)	1.88 (16.3)	1.77 (17.8)	−0.103 (−0.214, 0.007) [b]

Data is presented as geometric mean (CV%)

The ratios of the LSM and the 90% CIs of the AUC_{0-12}, $AUC_{0-\infty}$ and C_{max} for levodopa and carbidopa were within the range of 80% to 125%, indicating that once-daily dosing with 80 mg istradefylline for 14 days had no significant effect on the rate or extent of levodopa or carbidopa exposure when administered as a single dose of 200/50 mg L/C. The statistically significant 3-minute difference in $t_{1/2}$ between Days 1 and 15 was not considered to be clinically relevant.

Study PP15710 was a DB, placebo-controlled, randomised, multiple ascending dose study of istradefylline in elderly male and female subjects (age >55 years). The study evaluated the effect of smoking on

istradefylline PK (a secondary objective). The study was divided into 4 groups. Group 3 (6 male, 6 female non-smoking subjects) and Group 4 (8 male smoking subjects [at least 5 cigarettes/day for at least 1 year]) received the same dose of istradefylline 40 mg/day for 14 days.

The $t_{1/2}$ of istradefylline in non-smokers was up to 2.7-fold that in smokers, and the apparent clearance was ~3.1-fold lower in non-smokers than in smokers. On Day 14, the rate and extent of systemic exposure of non-smokers to istradefylline was significantly higher than that in smokers ($p \leq 0.013$). The mean ratios for C_{max} and AUC_{0-24} (smokers to nonsmokers) are presented in Table 27.

Table 27: Mean C_{max} and AUC_{24} Ratios (Smokers to Non-smokers) of Ro 64-4529 and their 95% CIs. Comparison of Group 3 males and Group 4

Parameter	Day	Ratio	Lower	Upper
C_{max}	1	0.94	0.63	1.40
	14	0.56	0.37	0.84
AUC_{24}	1	0.69	0.37	1.30
	14	0.37	0.20	0.69

Study 6002-US-016 was a phase 1, multicentre, OL, parallel-group study to evaluate the influence of moderate hepatic impairment (Child-Pugh B Classification) on the PK of istradefylline. The impact of smoking (>20 cigarettes/day) was evaluated as a secondary objective.

The PK parameters of istradefylline were significantly altered in smoking subjects by Day 14 following daily oral administration of 40 mg istradefylline. The mean AUC_{0-24}, AUC_{0-t} and $AUC_{0-\infty}$ of istradefylline were significantly decreased in smokers (hepatically impaired smokers and healthy smokers) compared with nonsmokers (hepatically impaired nonsmokers and healthy nonsmokers).

Results of the statistical analyses of the difference in istradefylline PK between smokers and nonsmokers are provided in Table 28 and Table 29. The LSM AUC_{0-t} ratio was 0.29 in hepatically impaired smokers compared to hepatically impaired nonsmokers and 0.21 in healthy smokers compared to healthy nonsmokers. The LSM $AUC_{0-\infty}$ ratio was 0.23 in hepatically impaired smokers compared to hepatically impaired nonsmokers, and 0.18 in healthy smokers compared to healthy nonsmokers.

Table 28: Statistical evaluation of the difference in Istradefylline PK parameters between hepatically impaired smokers and hepatically impaired nonsmokers

Parameter	Statistic	Hepatically Impaired Smoker	Hepatically Impaired Nonsmoker	Ratio[a,c]	90% CI[b]
AUC_{0-24} (ng·h/mL)	n	7	7		
	Mean	5183.50	8111.52	0.64	41.23, 99.03
$AUC_{0-\infty}$ (ng·h/mL)	n	7	6		
	Mean	22432.15	96802.41	0.23	8.87, 60.52
AUC_{0-t} (ng·h/mL)	n	7	7		
	Mean	21077.11	72607.91	0.29	12.72, 66.24
C_{max} (ng/mL)	n	7	7		
	Mean	351.68	489.26	0.72	48.63, 106.24
$t_{1/2}$ (hour)	n	7	6		
	Mean	99.63	287.61	0.35	-1.11, 70.39[d]

Note: For AUCs and C_{max} mean was evaluated by exponentiating the LSM of the $\log_e$ transformed PK parameter. For $t_{1/2}$ mean was the LSM on original scale. The LSM is from the ANOVA model.
a For AUCs and C_{max}, ratio and CIs are based on back log-transformation of the log-transformed data. For $t_{1/2}$, ratio and Cis are based on original data. To obtain percentage multiply value by 100.
b 90% CI for mean group differences was calculated from ANOVA. Limits of the Cis expressed as percentages
c To obtain percentage multiply value by 100.

d For t1/2 on original scale, lower limit of 90% CI was calculated by (Lower Estimate + LSM [Reference]) / LSM [Reference] 100% by converting a difference to a ratio scale based on the reference mean. When the lower estimate was negative and its absolute value was greater than the LSM of reference, the lower limit of the 90% CI was negative.

Table 29: Statistical evaluation of the difference in Istradefylline PK parameters between healthy smokers and healthy nonsmokers

Parameter	Statistic	Healthy Smokers	Healthy Nonsmoker	Ratio[a,c]	90% CI[b]
$AUC_{0\text{-}24}$ ng·h/mL)	n	7	7		
	Mean	5205.64	8913.93	0.58	37.68, 90.50
$AUC_{0\text{-}\infty}$ (ng·h/mL))	n	7	6		
	Mean	14981.03	84758.02	0.18	6.77, 46.16
$AUC_{0\text{-}t}$ (ng·h/mL)	n	7	7		
	Mean	14682.34	71088.61	0.21	9.05, 47.13
C_{max} (ng/mL)	n	7	7		
	Mean	379.72	479.11	0.79	53.62, 117.14
$t_{1/2}$ (hour)	n	7	6		
	Mean	55.55	117.99	0.47	-40.05, 134.22[d]

Note: For AUCs and C_{max} mean was evaluated by exponentiating the LSM of the log_e transformed PK parameter. For $t_{1/2}$ mean was the LSM on original scale. The LSM is from the ANOVA model.
a For AUCs and C_{max}, ratio and CIs are based on back log-transformation of the log-transformed data. For $t_{1/2}$, ratio and Cis are based on original data. To obtain percentage multiply value by 100.
b 90% CI for mean group differences was calculated from ANOVA. Limits of the Cis expressed as percentages
c To obtain percentage multiply value by 100.
d For t1/2 on original scale, lower limit of 90% CI was calculated by (Lower Estimate + LSM [Reference]) / LSM [Reference] 100% by converting a difference to a ratio scale based on the reference mean. When the lower estimate was negative and its absolute value was greater than the LSM of reference, the lower limit of the 90% CI was negative.

Based on the pop-PK model and PK-PD Percent OFF time model [6002-POP-PK-ANALYSIS (2006), a PD patient who smokes and receives a 20 mg/day dose would have a response to istradefylline that was 18 (12-26) % lower than a similar patient who does not smoke. For a 40 mg/day dose, in the same two patients, the smoker's response would be 12 (7.3 – 17) % lower. An increase in dose from 20 to 40 mg/day for the smoker would result in a predicted response that would be similar to the response demonstrated in a non-smoker who receives 20 mg/day. For an istradefylline response equivalent to the effect expected at 40 mg/day in non-smokers, smokers would require a dose of approximately 60 mg/day. The dose-response relationship is not directly proportional due to the non-linearity of the E_{max} exposure-response (E-R) model.

Exposure relevant for safety evaluation

In the thorough QTc study (6002-US-024), istradefylline was administered to healthy male and female subjects at 40 mg and 160 mg once daily for 14 days. The PK parameters for istradefylline are summarized in Table 30.

Table 30: Mean (SD) PK parameters of istradefylline on Days 1 and 14 after repeated administration of istradefylline 40 and 160m/day in healthy male and female subjects (Study 6002-US-024)

Parameter	40 mg (N = 44)	160 mg (N = 43)
Day 1		
T_{max} (h)[a]	3.00 (2, 6)	4.00 (1, 5)
C_{max} (ng/mL)	271.5 (60.6)	599.9 (147.4)
$AUC_{0\text{-}24}$ (ng·h/mL)	2462 (586)	6879 (1603)
Day 14		
T_{max} (h)[a]	3.00 (0, 16)	4.00 (0.5, 24)
C_{max} (ng/mL)	518.4 (171.2)	1556.9 (516.8)
$AUC_{0\text{-}24}$ (ng·h/mL)	9228 (3233)	29279 (9263)

a Median (range)

The exposure ratio between the active M1 metabolite and istradefylline was less than 5% on Day 1 and Day 14. On Day 14, the mean AUC_{0-24} of the M1 metabolite was 201.12 ng·h/mL for the 40 mg dose and 475.66 ng·h/mL for the 160 mg dose. Mean C_{max} values were 11.42 ng/mL and 26.78 ng/mL for the 40 and 160 mg doses, respectively.

2.4.3. Pharmacodynamics

Adenosine A_{2A} receptor occupancy (RO) in the human brain, a thorough QT/QTc study and an abuse potential study. Three PK-PD analyses (efficacy and safety) were conducted.

Mechanism of action

Istradefylline is a selective adenosine A_{2A} receptor antagonist. Among four adenosine receptor subtypes (A_1, A_{2A}, A_{2B} and A_3) within the human central nervous system, the adenosine A_{2A} receptor is located almost exclusively in the basal ganglia. There is evidence that A_{2A} receptors located on the striatopallidal medium spiny neurons in the indirect pathway are involved in motor control via the basal ganglia. It is hypothesized that blockage of the A_{2A} receptors by istradefylline reduces the excitability of this indirect pathway that occurs in PD, resulting in an improvement in PD symptoms.

Primary pharmacology

Study 6002-EU06 was a phase 1, OL, multiple dose, clinical pharmacology study in 12 healthy men (aged 35 to 65 years), using [^{11}C]-istradefylline and Positron Emission Tomography (PET) to investigate adenosine A_{2A} RO by istradefylline in the human brain.

Two subjects received a single intravenous dose of 300 MBq radiolabelled [^{11}C]-istradefylline prior to PET scan. Ten subjects received once daily oral dosing with unlabelled istradefylline for 14 days. Istradefylline doses were 40, 20, 5, 1.5, 0.5, and 0.1 mg; two subjects per dose level, except 1.5 mg and 0.1 mg dose levels with one subject each. Istradefylline plasma concentrations were above the lower limit of quantification (LLOQ) following all doses and there was a dose-related trend in concentrations.

PET showed that over 90% RO was achieved with daily doses of greater than 5 mg istradefylline for 14 days, which did not allow the construction of a dose-occupancy curve. Dosing with 40 mg, 20 mg and 5 mg daily for 14 days achieved almost complete suppression of the binding of radiolabelled istradefylline. At doses of 5 mg and below, RO decreased proportionally and construction of a dose-occupancy curve was possible.

The radiolabelled istradefylline kinetic PET data were best described by a two-tissue compartmental model with a blood volume component, which allowed estimation of the total tissue volume of distribution and binding potential. After applying an appropriate saturation kinetic model, which included a term for non-specific binding, the half saturation dose (ED_{50}) for istradefylline for each region of interest estimated from total tissue volume of distribution was 0.55 mg for caudate, 1.28 mg for cerebellum, 0.49 mg for putamen, 0.47 mg for thalamus, and 0.30 mg for nucleus accumbens.

Secondary pharmacology

Study 6002-US-024 was a thorough QTc study to examine the effect of istradefylline on ECG parameters. This was a single-center, Phase 1, double-blind (except for the use of moxifloxacin as a positive control), randomised, parallel-group study. 175 non-smoking, healthy subjects (50% male, 50% female), aged between 18 and 45 years, received multiple oral doses of placebo or istradefylline at a dose of 40 or 160 mg/day once daily for 14 days.

There was no evidence that istradefylline affected heart rates and there were no bradycardic outlier imbalances between istradefylline and placebo. There were 40% tachycardic outliers at Day 14 in the 40 mg istradefylline dose group versus 15% in the placebo group. The reason for this tachycardic finding is unknown, but does not appear to be drug-related since the effect was similar for 160 mg istradefylline (21%) and placebo (15%). There was no evidence that istradefylline changed PR and QRS conduction intervals. No PR or QRS outliers were observed.

The placebo-corrected time-averaged mean change from Baseline for moxifloxacin was 4.5 millisecond on Day 1 and a 6.2 millisecond on Day 14. Thus, assay sensitivity at the 5 millisecond level was observed. No effect of istradefylline on cardiac repolarisation was observed. At istradefylline doses of 40 mg and 160 mg, the placebo-corrected change from Baseline QTcI effect was -1.7 milliseconds and -0.3 milliseconds on Day 1, respectively, and -1.7 and -0.2 milliseconds on Day 14, respectively.

The 90% upper CIs for the placebo-corrected change from baseline based on the time-matched analysis for QTcI on Day 14 are shown in the below table. Istradefylline 160 mg showed an upper CI that did not cross 10 milliseconds except at 5, 6 and 12 hour timepoints, when the upper bound was exceeded by 0.4, 0.3 and 0.01 milliseconds, respectively. Istradefylline 40 mg showed an upper CI <10 milliseconds at all timepoints. The applicant considered that the greater variability of the time-matched analysis and the fact that this study had small sample sizes for each group may account for this finding.

Table 31: 90% Upper CIs (ms) for the placebo-corrected change from baseline: time-matched analysis on Day 14

Time Point (h)	Istradefylline 40 mg	Istradefylline 160 mg	Moxifloxacin 400 mg
0.5	**4.39**	**6.22**	7.02
1	**5.66**	**5.47**	11.05
2	**5.64**	**6.04**	14.81
3	**5.97**	**8.00**	18.37
4	**9.25**	**8.39**	18.34
5	**6.28**	**10.39**	15.27
6	**4.63**	**10.27**	18.56
8	**1.18**	**3.22**	10.56
10	**4.20**	**5.55**	11.84
12	**6.37**	**10.01**	15.62
14	**7.75**	**8.21**	13.37
16	**5.71**	**7.16**	14.60
18	**6.07**	**7.12**	11.16
20	**7.25**	**6.92**	10.78
24	**7.38**	**8.92**	12.99

Using either the specific outlier criteria of a new >500 milliseconds QTcI duration, a change from Baseline of >60 milliseconds, or new abnormal U waves, no subject in either istradefylline group had these findings either after a single dose or at steady state (Day 14). Even the nonspecific criterion of a 30-60 milliseconds change from Baseline showed no difference in the number of subjects with this finding when istradefylline was compared with placebo.

Study 6002-017 was a DB, placebo- and active-controlled 6-way crossover study to evaluate the abuse potential of single doses of istradefylline compared to placebo and phentermine in recreational stimulant users. Subjects participated in a Screening visit, a 5-day Qualification (Drug Discrimination) Phase, a 6-period Treatment Phase, and a safety Follow-up visit. A total of 94 subjects were included in the Qualification Population. Of these, 55 (58.5%) completed the Qualification Phase and were randomised

in the Treatment Phase. A total of 42 (76.4%) subjects completed all 6 Treatment Periods and had a valid Drug Liking VAS Emax in each Treatment Period (Completer Population).

Primary endpoint: Maximum effect (E_{max}) of the Drug Liking Visual Analog Scale (VAS)

Results for the bipolar Drug Liking VAS E_{max} primary endpoint are provided in Table 32.

Table 32: Analysis Results for "At this moment" Drug Liking VAS E_{max}-Primary endpoint (completer population)

Pairwise Comparison	Mean Test	Mean Reference	Mean Difference (Test-Reference)	95% CI of Difference
Study Validity: Phentermine – Placebo				
Phen 45 mg – Placebo	72.02	57.90	14.12	**8.92, 19.32**
Phen 90 mg – Placebo	79.12	57.90	21.21	**16.06, 26.37**
Absolute Abuse Potential: Istradefylline – Placebo				
Istra 40 mg – Placebo	62.02	57.90	4.12	**0.72, 7.51**
Istra 80 mg – Placebo	62.74	57.90	4.83	**0.64, 9.03**
Istra 160 mg – Placebo	61.71	57.90	3.81	**0.82, 6.80**
Relative Abuse Potential: Istradefylline – Phentermine				
Istra 40 mg – Phen 45 mg	62.02	72.02	-10.00	**-14.91, -5.09**
Istra 40 mg – Phen 90 mg	62.02	79.12	-17.10	**-22.03, -12.16**
Istra 80 mg – Phen 45 mg	62.74	72.02	-9.29	**-14.62, -3.95**
Istra 80 mg – Phen 90 mg	62.74	79.12	-16.38	**-22.52, -10.24**
Istra 160 mg – Phen 45 mg	61.71	72.02	-10.31	**-14.64, -5.98**
Istra 160 mg – Phen 90 mg	61.71	79.12	-17.40	**-22.26, -12.55**

Drug Liking VAS item "At this moment, my liking for this drug is" where 0 = strong-disliking. 50-neither like nor dislike and 100=Strong liking.

Study validity was demonstrated by the statistically significant difference between phentermine and placebo on the Drug Liking VAS E_{max} primary endpoint. In terms of absolute abuse potential, there were statistically significant differences between istradefylline doses and placebo on the Drug Liking VAS E_{max}; however, these were considered not clinically meaningful as the upper limits of the 95% CIs of the differences at all dose levels were less than 11 (Chen & Bonson, 2013), with mean differences on the primary endpoint ranging from approximately 4 to 5 points on a 100-point scale vs. ~ 14 and 21 points with phentermine 45 mg and 90 mg, respectively. With respect to relative abuse potential, all 3 istradefylline doses showed statistically significantly lower effects relative to phentermine on the primary endpoint.

Secondary endpoints

There were some statistically significant differences between istradefylline and placebo on secondary endpoints of balance of effects, positive effects, stimulant effects, and any effects; however, the differences were relatively small compared to those observed both phentermine 45 mg and 90 mg. None of the istradefylline doses showed statistically significant negative effects compared to placebo. In addition, all 3 istradefylline doses were associated with median scores <10.0 on all Drug Similarity VAS, indicating that the majority of the subjects did not rate istradefylline as being similar to drugs of abuse or other mild stimulants, such as nicotine and caffeine, that they had previously experienced (Table 33).

Table 33: Summary of results for drug similarity VAS – Secondary Endpoint (Completer Population)

Drug/Drug Class	Statistic	Istra 40 mg N=42	Istra 80 mg N=42	Istra 160 mg N=42	Phen 45 mg N=42	Phen 90 mg N=42	Placebo N=42
Placebo	n	42	42	42	42	42	42
	Median (Min, Max)	0.0 (0, 100)	0.0 (0, 100)	0.0 (0, 100)	0.0 (0, 100)	0.0 (0, 100)	88.0 (0, 100)
Caffeine	n	38	38	38	38	38	38
	Median (Min, Max)	1.5 (0, 87)	0.0 (0, 87)	3.0 (0, 86)	13.0 (0, 100)	20.5 (0, 100)	0.0 (0, 75)
Cocaine[a]	n	33	33	33	33	33	33
	Median (Min, Max)	0.0 (0, 89)	0.0 (0, 100)	2.0 (0, 96)	17.0 (0, 86)	25.0 (0, 90)	0.0 (0, 43)
Amphetamines[b]	n	37	37	37	37	37	37
	Median (Min, Max)	1.0 (0, 100)	8.0 (0, 99)	2.0 (0, 92)	36.0 (0, 99)	59.0 (0, 100)	0.0 (0, 100)
Nicotine	n	31	31	31	31	31	31
	Median (Min, Max)	0.0 (0, 32)	0.0 (0, 49)	0.0 (0, 41)	0.0 (0, 47)	0.0 (0, 57)	0.0 (0, 20)
Ecstasy (MDMA)	n	33	33	33	33	33	33
	Median (Min, Max)	0.0 (0, 77)	0.0 (0, 48)	0.0 (0, 95)	8.0 (0, 85)	23.0 (0, 100)	0.0 (0, 37)
Opioids[c]	n	29	29	29	29	29	29
	Median (Min, Max)	0.0 (0, 58)	0.0 (0, 11)	0.0 (0, 24)	0.0 (0, 37)	0.0 (0, 25)	0.0 (0, 37)
LSD	N	18	18	18	18	18	18
	Median (Min, Max)	0 (0, 0)	0 (0, 21)	0 (0, 0)	0 (0, 0)	0 (0, 27)	0 (0, 6)
THC[d]	n	41	41	41	41	41	41
	Median (Min, Max)	0.0 (0, 33)	0.0 (0, 37)	0.0 (0, 62)	0.0 (0, 80)	0.0 (0, 65)	0.0 (0, 36)

Drug Similarity VAS item "How similar is the drug you most recently received to [drug name]?" where 0=Not at all similar and 100=Very similar
A Including "crack"
B d-Amphetamine and "meth"
C Morphine, Oxycodone and hydrocodone
D Marijuana, cannabis, hash
Note Benzodiazepine, Ketamine and phencyclidine data are not shown as the sample size were very small (≤ 5 subjects).
Istra= Istradefylline; LSD = Lysergic acid diethylamide, MDMA = 3, 4-methylenedioxymethamphetamine; Phen=Phentermine, THC= Tetrahydrocannabinol; VAS= Visual analogue Scale

With respect to relative abuse potential, all 3 istradefylline doses showed statistically significantly lower effects relative to phentermine on the primary endpoint of Drug Liking VAS E_{max} and the majority of secondary endpoints of balance of effects (Drug Liking VAS TA-AUE at 40 mg and 80 mg, Overall Drug Liking VAS E_{max}, Take Drug Again VAS E_{max}), positive effects (Good Effects VAS Emax and TA-AUE), stimulant effects (Alertness/Drowsiness VAS E_{max}, Agitation/Relaxation VAS E_{max}), negative effects (Bad Effects VAS E_{max} and TA-AUE), and any effects (Any Effects VAS E_{max} and TA-AUE), particularly when compared to the 90 mg phentermine dose. Although a few endpoints did not show statistically significant differences, this was primarily with the 80 mg and 160 mg doses of istradefylline (i.e., Drug Liking VAS TA-AUE [istradefylline 160 mg vs. phentermine 90 mg], Overall Drug Liking VAS Emax [istradefylline 80 mg, and 160 mg vs. phentermine 45 mg], Good Effects VAS TA_AUE [istradefylline 80 mg, and 160 mg vs. phentermine 45 mg], Alertness/Drowsiness VAS TA_AUE [istradefylline 40 mg, 80 mg, and 160 mg vs. phentermine 90 mg], Agitation/Relaxation VAS Emax [istradefylline 40 mg, 80 mg, and 160 mg vs. phentermine 45 mg]).

Pharmacokinetics

Sparse PK samples were obtained to confirm exposure to istradefylline at predose, 0.5, 1, and 3 hours post-dose. Peak plasma concentrations were observed at the 3-hour sampling timepoint, with mean (SD) istradefylline concentrations of 146.76 (42.517) ng/mL, 209.33 (67.708) ng/mL, and 298.59 (106.925) ng/mL for the 40 mg, 80 mg, and 160 mg doses, respectively.

Pharmacodynamic interactions

The potential for istradefylline to have PD interactions with other anti-Parkinson's drugs is low. Istradefylline has a different pharmacological target from other anti-Parkinson's drugs presenting high selectivity/specificity to the A_{2A} receptors relative to other receptors in human brain.

Genetic differences in PD response

Although formal studies examining the effect of genetic differences on the PD response to istradefylline have not been conducted, the association of genetic polymorphisms within the adenosine A_{2A} receptor was examined in the literature. Based on this evidence, it does not appear that polymorphism affects the response to istradefylline.

Relationship between plasma concentration and effect

6002-pop-pk-analysis (2006): Pop-PK-PD analysis of istradefylline in healthy subjects and in patients with PD

Data from 6 Phase 2/3 clinical trials comprised the final PK-PD database. The Phase 2/3 clinical trials included multiple dose data from patients with PD experiencing motor complications being treated with L/C.

Exposure-efficacy analysis

- Percent OFF Time

The relationship between istradefylline AUC_{ss} and Percent OFF time was best described by a non-linear PK-PD model based on time for the disease progression/placebo response (DP-PR) component and an Emax model for the effect of istradefylline, which entered the model as additive to the DP-PR component. The estimated population maximum decrease in Percent OFF time due to istradefylline exposure was an additional 5.79% (4.10-7.50%) above the placebo effect. The most influential covariates on the Percent OFF time PK-PD model were smoking status (through effects on PK exposure) and the presence of COMT inhibitors (mediated as PD effect).

- Probability of ON Time Dyskinesia

The relationship between istradefylline AUC_{ss} and the probability of ON time dyskinesia was best described by a linear slope-intercept model based on time for the DP-PR component and a linear slope model based on the normalized AUC for the effect of istradefylline, which entered the model as additive to the DP-PR component. There was a linear decrease in the probability of ON time dyskinesia as a result of being in the study (DP-PR portion of model) and a linear decrease in the probability of ON time dyskinesia with exposure to istradefylline. The naive-pooled population model-predicted typical probability of ON time dyskinesia after 42 days of the study was 47.7%, 46.6%, 45.4%, 44.7%, respectively, for the median exposures associated with the placebo, 20 mg/day, 40 mg/day, and 60 mg/day dose groups. This represents an approximate decrease of 1% in the probability of ON time dyskinesia for every 20 mg/day of istradefylline.

- Percent ON Time with Dyskinesia

The relationship between istradefylline AUC_{ss} and Percent ON time with dyskinesia was best described by a DP-PR component that incorporated time as an exponential function and a linear slope effect for istradefylline, which entered the model as additive to the DP-PR component. The estimated population slope for the E-R relationship was 2.50% per 5000 ng*hr/mL of istradefylline exposure. Since istradefylline exposure results in an increase in ON time, it is likely that the relationship demonstrated between istradefylline exposure and Percent ON time with dyskinesia is reflecting the general increase in ON time rather than a specific increase in ON time with dyskinesia.

- UPDRS subscale 3 score PK-PD model

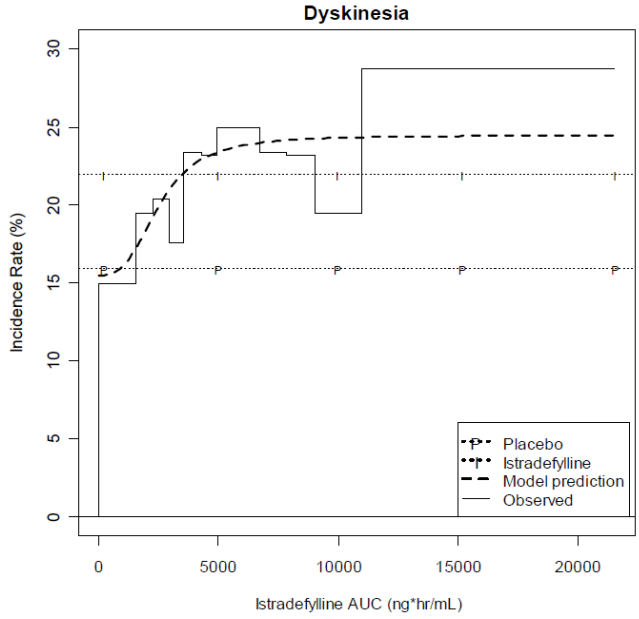
The relationship between istradefylline AUCss and Unified Parkinson's Disease Rating Scale (UPDRS) subscale 3 score was best described by an Emax model based on time for the DP-PR component and a linear slope effect for istradefylline, which entered the model as additive to the DP-PR component. The estimated population slope for the E-R relationship was -0.206% per 5000 ng*hr/mL of istradefylline exposure. At 60 mg/day, the maximum typical change in UP30 due to istradefylline, would be -1.12%.

Exposure-safety analysis

The relationships between exposure (popPK individual predicted istradefylline AUC_{ss}) and AE were investigated graphically, and by using logistic regression models for naive-pooled dichotomous outcome data. Any AE that occurred with an incidence that was 2% or higher than placebo and demonstrated an E-R trend graphically was subject to formal modelling using logistic regression. Based on this review, dyskinesia, dizziness, and nausea AE were selected for E-R modelling.

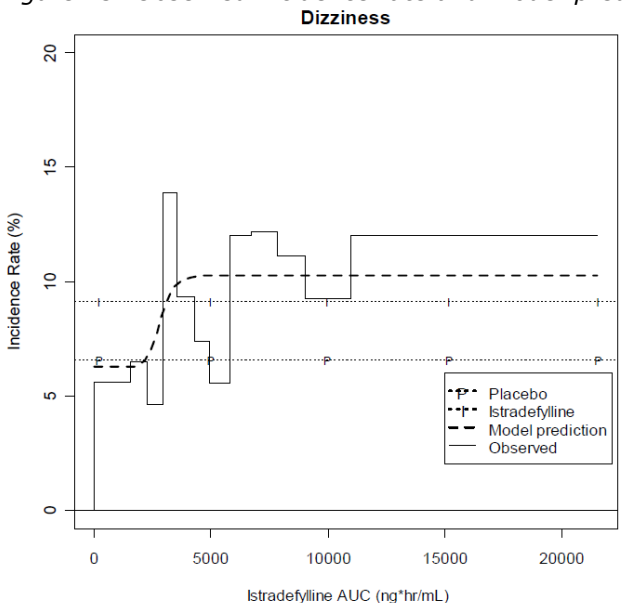
The relationship between istradefylline AUC_{ss} and the probability of experiencing dyskinesia as an AE was best described by a sigmoid E_{max} model. A plot of observed dyskinesia incidence vs. predicted is presented in Figure 14.

Figure 14: Observed incidence rate and model-predicted probability of dyskinesia vs istradefylline AUCss



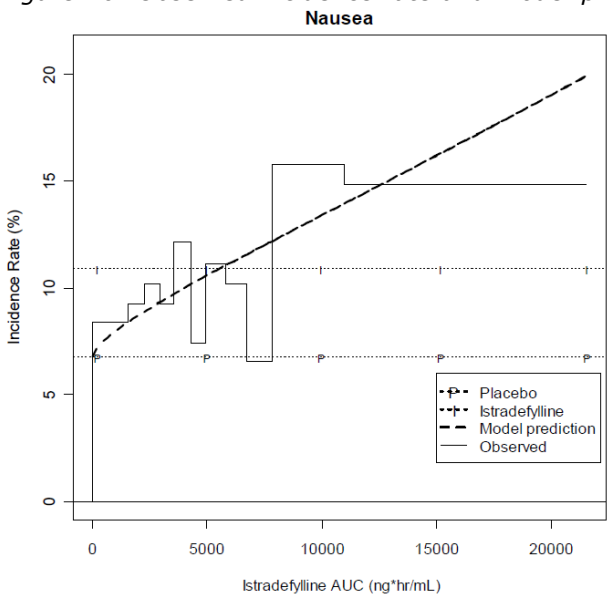
The relationship between istradefylline AUC_{ss} and the probability of experiencing dizziness as an AE was best described by a sigmoid Emax model. A plot of observed dizziness incidence vs. predicted is presented in Figure 15.

Figure 15: Observed incidence rate and model-predicted probability of dizziness vs. istradefylline AUC_{ss}



The relationship between istradefylline AUC_{ss} and the probability of experiencing nausea as an AE was best described by a power model. A plot of observed dizziness incidence vs. predicted is presented in Figure 16.

Figure 16: Observed incidence rate and model-predicted probability of nausea vs. istradefylline AUCss



Evaluation of doses relative to the main clinical endpoints for efficacy and safety

Simulations based on the popPK-PD models showed that efficacy, as measured by the maximum change in Percent OFF time or the percent of patients with a decrease in OFF time of 30 min or more begins to plateau at doses greater than 40 mg/day. The plateau for the probability of dyskinesia and dizziness is also reached at 40 mg/day. The probability of nausea will not reach a plateau but would be expected on average to increase in probability by approximately 1.5-2% for every 20 mg/day increase in the dose of istradefylline above 20 mg/day.

Table 34: Integrated Percent OFF time and Dyskinesia, Dizziness and Nausea probability by dose

Dose (mg)[b]	Change in Percent OFF Time Endpoint (%)	Change in Actual OFF Time Endpoint (hr)	%Patients with >=30 min Decrease in Actual OFF Time Endpoint	Dyskinesia Probability	Dizziness Probability	Nausea Probability
5	-2.2 (-2.8, -1.6)[a]	-0.35 (-0.45, -0.25)	57.5	16.3 (13.4, 20.9)	6.3 (4.8, 8.9)	8.2 (6.5, 10.8)
10	-3.2 (-4.1, -2.3)	-0.51 (-0.66, -0.37)	61.5	18.6 (15.0, 24.2)	6.7 (5.1, 9.5)	8.9 (7.1, 11.3)
20	-4.0 (-5.1, -2.9)	-0.64 (-0.81, -0.46)	64.7	22.5 (18.6, 25.4)	9.3 (6.1, 11.9)	9.9 (7.9, 12.1)
40	-4.7 (-6.0, -3.4)	-0.75 (-0.96, -0.54)	67.3	24.1 (21.0, 27.5)	10.7 (8.6, 13.2)	11.8 (9.5, 14.1)
60	-4.9 (-6.3, -3.6)	-0.79 (-1.01, -0.57)	68.4	24.3 (21.3, 28.1)	10.9 (8.6, 14.1)	13.0 (10.6, 16.2)
80[c]	-5.1 (-6.6, -3.7)	-0.82 (-1.04, -0.59)	68.9	24.4 (21.3, 30.0)	11.0 (8.7, 15.8)	15.2 (11.1, 20.1)

a median (95% CI)
b median AUC at each dose and bootstrap results used to calculate endpoint data
c 80mg dose was extrapolated using 60mg data.

6002-014-pkpd-r-en (2017): E-R analysis of istradefylline

Data from the phase 3 study 6002-014, which evaluated subjects with advanced PD who were already maximally/optimally treated with approved therapies, were included in this analysis. The PK-PD dataset included 593 subjects with both efficacy and PK data, and 610 subjects with both safety and PK data.

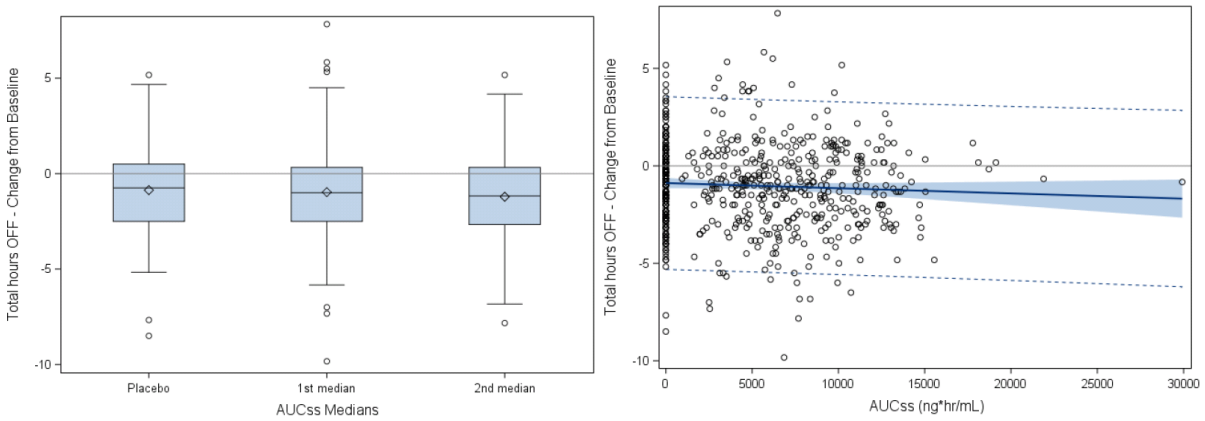
ER relationships were evaluated using istradefylline exposure (AUC_{ss}) as a categorical variable for ANOVA and as a continuous variable for linear regression, which was predicted from the population PK model 6002-014-pop-pk-r-en (2017).

Exposure-efficacy analysis

- Total hours per day spent in OFF state

The mean values of T_{OFF} change from baseline at Week 12 were not significantly different among istradefylline AUCss medians or individual AUCss values (Figure 17).

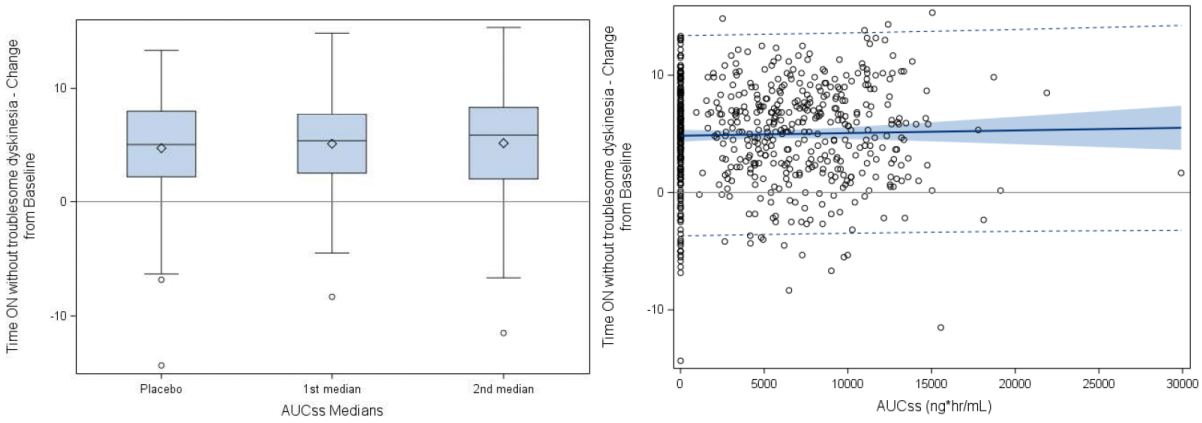
Figure 17: OFF time E-R by AUCss median (left) and individual AUCss values (right)



- Total Hours of ON Time per Day Without Troublesome Dyskinesia

The mean values of T_{ON} change from baseline at Week 12 were not significantly different among istradefylline AUCss medians or individual AUCss values (Figure 18).

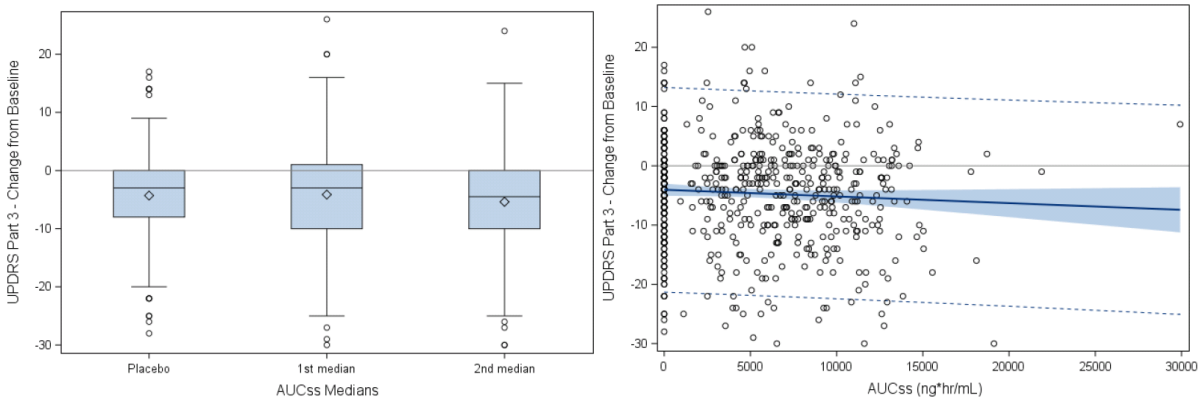
Figure 18: ON time E-R by AUCss median (left) and individual AUCss values (right)



- UPDRS motor examination score (Part 3)

The mean values of UPDRS motor examination score (Part 3) change from baseline at Week 12 were not significantly different among istradefylline AUC_{ss} medians or individual AUC_{ss} values (Figure 19).

Figure 19: UPDRS motor examination score (Part 3) E-R by AUCss median (left) and individual AUCss values (right)

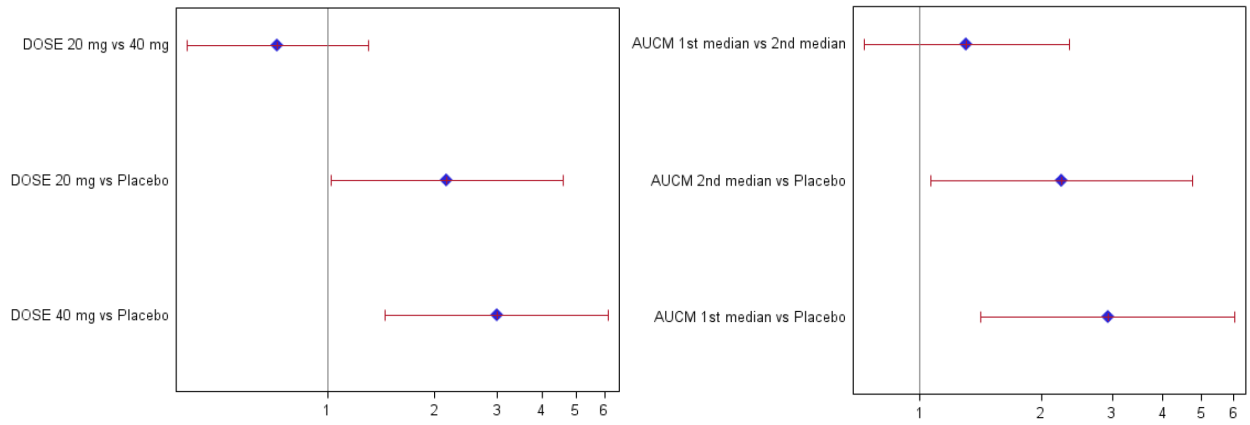


Exposure-safety analysis

The AE reported with the highest frequencies were dyskinesia (10.3%), falls (7.2%) and nausea (3.8%).

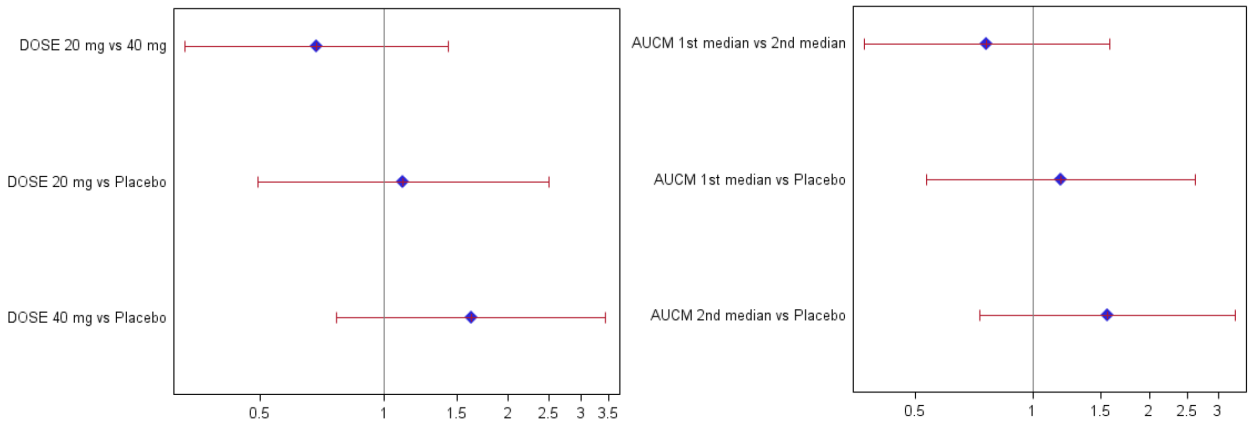
Dyskinesia frequency increased with increasing exposure, with a significantly higher odds ratio at higher levels of exposure whether measured by dose or AUCss median (Figure 20).

Figure 20: Dyskinesia E-R odds ratios by dose (left) and AUCss median (right)



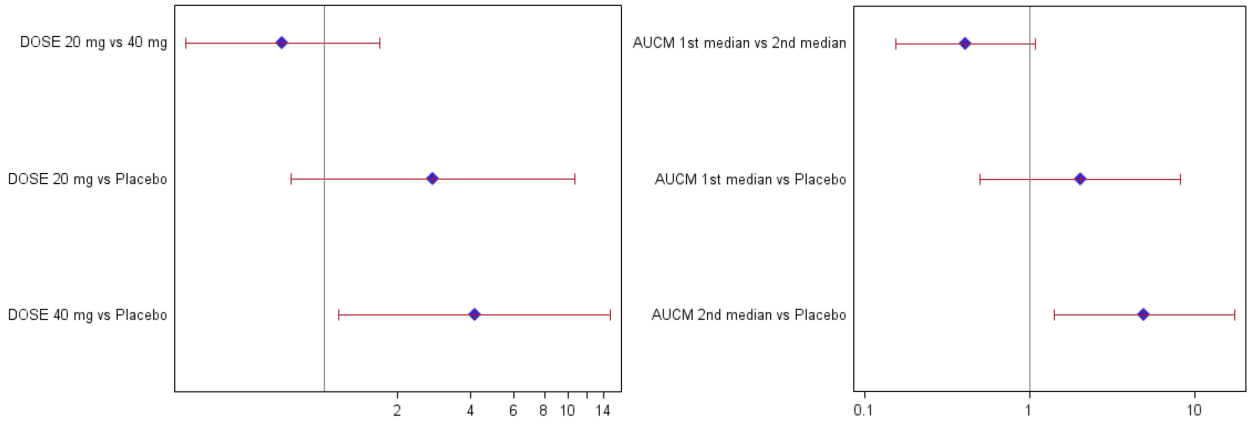
The frequency of falls increased with increasing exposure but the increased risk was not significant (Figure 21).

Figure 21: Falls E-R odds ratios by dose (left) and AUCss median (right)



The frequency of nausea increased with increasing exposure, with a significantly higher odds ratio for the AUCss upper median when compared to placebo (Figure 22).

Figure 22: Nausea E-R odds ratios by dose (left) and AUC_{ss} median (right)



Constipation showed a trend of increasing incidence with increased exposure. Hypotension exhibited increased frequency for the first median relative to placebo, but the increased risk was not significant. Dizziness, somnolence, and hallucinations exhibited no clear E-R relationship. The maximum reported total score for parts A through D (gambling, sex, buying, eating) of the Questionnaire for Impulsive-Compulsive Disorders in Parkinson's Disease - Rating Scale (QUIP-RS) exhibited no trends with increasing AUCss median.

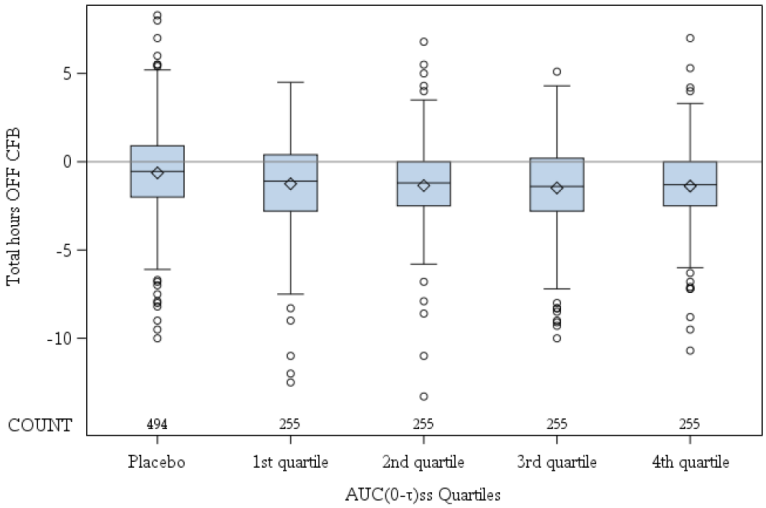
6002-nda-response-E-R-analysis-r-en (2018): E-R analysis of istradefylline

Data from 5 phase 2/3 clinical studies (6002-US-005, 6002-US-006, 6002-US-013, 6002-0608, and 6002-009), which evaluated subjects with moderate to severe PD, were included in this analysis. The final PK-PD dataset included 1514 subjects: 1020 randomised to active treatment and 494 randomised to placebo.

E-R relationships were evaluated using categorical exposure metrics predicted from the pop-PK model 6002-nda-response-pop-pk-r-en (2018).

Exposure-efficacy analysis

Figure 23: TOFF change from baseline at Week 12 by AUC(0-τ)ss quartile



Week 12 TOFF change did not demonstrate an E-R relationship across the 20 to 60 mg dose range. However, all exposure quartiles in active treatment groups across the 20 to 60 mg dose range were significantly different from placebo.

Exposure-safety analysis

No apparent E-R relationships or trends were found for the incidence of AEs investigated, except for an increase in liver enzyme elevations with increasing exposure from 0.8% in the lowest quartile of exposure to 4.7% in the highest quartile of exposure. There were no serious liver enzyme elevations.

2.4.4. Discussion on clinical pharmacology

Pharmacokinetics

Analytical methods

Bioanalytical Method Validation

Most validation reports were generated prior to the establishment of current EMA recommendations on bioanalytical method validation (EMEA/CHMP/EWP/192217/2009 Rev. 1 Corr. 2**). Therefore, not all methods have been fully validated to current expectations. Notwithstanding the inconsistent submission of certain data for specific validation parameters, the methods utilised to determine istradefylline and associated metabolites during clinical development are generally acceptable. Methods used to generate data for pop-PK analyses, investigate DDI, and PK in special populations were appropriate. While some PK studies utilised methods that were not fully validated according to current standards, the totality of evidence from additional PK investigations using fully validated methods is sufficient and provides supporting evidence.

Analytical Run Validation and Sample Analysis

The accuracy and precision for calibration standards and QC samples used during the analysis of participant samples was generally acceptable.

Because most clinical studies were performed before guidelines on the necessity to perform ISR were issued, ISR was not routinely performed. Consequently, the reproducibility of data cannot be guaranteed. However, given the accuracy and precision of calibration and QC samples was generally acceptable, this provides some reassurance regarding sample analysis. The majority of samples were analysed within the validated storage period. The atorvastatin and its metabolites determination in Study 6002-US-020 are considered reliable by the applicant based on stability of QCs used in the analysis (44 days at -20°C) and based on literature data in a similar method (human EDTA plasma) all covering a period longer than 66 days. The justification is considered acceptable.

Pop-PK analyses

6002-pop-pk-analysis (2006): Pop-PK analysis of istradefylline in healthy subjects and in patients with PD

The methods used for model development and evaluation are generally acceptable. A 2-compartment model with linear clearance and first order absorption provided an adequate fit to the data. Covariate effects included in the final model were Lean Body Mass (LBM), age, Race, concomitant CYP3A4 inhibitors, and smoking status on CL/F; Weight on V2/F; LBM on Q/F; Weight on V3/F; and food status on F. However, only the effects of smoking and the presence of CYP3A4 inhibitors as concomitant medications were deemed important in terms of clinical relevance.

Final model estimates for the structural and random variance parameters demonstrated good precision while some of the covariate effects were estimated with poor precision. The addition of covariate effects resulted in a minimal reduction (~1-2%) of interindividual variability in V/F and CL/F, when compared to the base model. Shrinkage of apparent clearance was low and individual predicted AUC, used in subsequent PK-PD analyses, can be considered reliable. The model is deemed fit for its intended purpose.

6002-014-pop-pk-r-en (2017): Pop-PK analysis of istradefylline in patients with PD

Appropriate methods were used for model development and evaluation. Data exclusions were well documented and are acceptable. The final model was a two-compartment model with linear clearance and three absorption transit compartments absorption. Interindividual variability was high for all parameters and largely unexplained by covariate effects. Covariate effects included in the final model were BMI and sex on CL/F, and body weight on V1/F. The effects for weight on V2/F and BMI on CL/F demonstrated a pronounced change in parameter values over the ranges of these covariates. However, the model predicts that CL/F decreases and V/F increases with increasing body size, which is not plausible. The effect of sex on CL/F was relatively modest and likely driven by differences in body size.

There were no apparent effects of race, concomitant anti-Parkinson's medications, caffeine use, mild hepatic, or mild/moderate renal impairment on istradefylline PK. The number of smoking subjects,

subjects with moderate hepatic or severe renal impairment and non-white subjects was too small to allow definitive evaluation.

All final PK model parameters were estimated with reasonably good precision (all SE <30%). The observed vs population and individual predicted concentration plots showed some divergence from the line of unity at high concentrations and deficiencies were noted in the VPCs. However, attempts to improve model fit were unsuccessful. Overall, the model is deemed reasonable given the sparse data sampling and large number of patients included in the analysis.

6002-nda-response-pop-pk-r-en (2018): Pop-PK analysis of istradefylline in patients with PD

Appropriate methods were used for model development and evaluation. Data exclusions were well documented and are acceptable. A 2-compartment model with linear clearance and first order absorption provided an adequate fit to the data. Interindividual variability was high for all parameters and largely unexplained by covariate effects. Covariate effects included in the final model were weight, Asian race, sex and bilirubin on CL/F, and weight on V1/F.

All final PK model parameters were estimated with good precision except for Q/F which was poorly estimated. Overall, the goodness-of-fit plots showed reasonable concordance between predicted and observed concentrations, and the VPC plots generally showed that the predictive ability of the final model was adequate. ETA shrinkage was low for CL/F (15.3%) but high for V1/F (60.3%). Therefore, clearance-based exposure estimates used in subsequent ER analyses, such as AUC, can be considered reliable.

The final model suggests CL/F decreases with increasing weight, which is not plausible. This is possibly due to the inclusion of sex and race effects on CL/F, which may be correlated with body weight. No relationship between hepatic function and smoking status with istradefylline clearance was identified. However, the majority of patients included in this analysis had normal hepatic function (95%) and were nonsmokers (95%). Bilirubin level was identified as a significant covariate and showed a negative relationship with CL/F. However, the effect was small and unlikely to be of clinical relevance.

Bioavailability

Single dose PK of istradefylline in healthy volunteers

Study 6002-9601 investigated daily doses of 10-200 mg under fasting conditions. Istradefylline C_{max} and $AUC_{0-\infty}$ both increased in a dose dependent manner, ranging from 43.0 to 342.3 ng/mL and 822.2 to 12240.6 ng*h/mL, respectively. t_{max} was observed at 2.5 to 3.5 hours.

Study 6002-EU01 investigated daily doses of 5-400 mg under fasting conditions. Istradefylline C_{max} and AUC_{0-72} both increased in a dose dependent manner, ranging from 16.6 to 507.1 ng/mL and 302.1 to 19949 ng·h/mL, respectively. t_{max} was observed between 2 to 5 hours.

Multiple dose pharmacokinetics of istradefylline in healthy volunteers

In Study 6002-US-002, plasma steady-state concentrations were achieved within 14 days following repeated administration of istradefylline at daily doses of 40, 60 and 80 mg.

In Study 6002-EU02, repeated dosing of istradefylline at 25 mg/day for 14 days resulted in an accumulation ratio for AUC_{24} of 5.6. Steady-state plasma concentrations were achieved after approximately 8 days of repeated dosing.

There was an apparent gender-specific difference in the terminal rate constant and corresponding $t_{1/2}$. Due to the small sample, no meaningful inference can be made based on the current data regarding gender-specific differences in $t_{1/2}$.

In Study 6002-9703, plasma steady-state concentrations were approximately achieved by Day 14 following repeated administration of istradefylline 20 mg/day.

In Study 6002-0104, plasma steady-state concentrations were generally achieved by Day 14 following administration of istradefylline at daily doses of 20, 40 and 80 mg.

Bioequivalence

In Study 6002-US-022, bioequivalence was demonstrated for the final commercial istradefylline formulation (Batch C4J0261), and previously administered tablets utilised during Phase 2b and 3 clinical trials (Batch F0946001 and Batch C3K0019, respectively).

In Study 6002-012, bioequivalence was demonstrated for the administration of two 10 mg istradefylline tablets compared to one 20 mg tablet.

Influence of food

In Study 6002-9601, under fed conditions, C_{max} was significantly increased by approximately 39% relative to fasting conditions following ingestion of 50 mg istradefylline. Feeding did not impact t_{max} or $AUC_{0-\infty}$.

In Study 6002-US-023, following the ingestion of a high-fat meal prior to administration of a 40 mg istradefylline, C_{max} and $AUC_{0-\infty}$ increased by 64% and 25%, respectively. Median t_{max} was decreased by approximately 1 hour in the presence of food for both istradefylline and metabolite M1, suggesting a faster rate of absorption under fed conditions.

In Study 6002-011, istradefylline C_{max} and AUC_{0-t} values were approximately 10 to 20% higher compared to fasting conditions following consumption of a low fat meal.

Overall, it is agreed that the impact of food on istradefylline PK is unlikely to be of clinical relevance.

Elimination

In the mass-balance study (6002-US-010), there was sufficient radioactivity recovery, amounting to 86.89 ± 8.61 % of the total dose (38.90 ± 10.70% and 47.99 ± 13.36% recovered in urine and faeces, respectively). Unchanged istradefylline was not detected in the urine which suggests that renal elimination is a minor excretion pathway.

As istradefylline is hepatically cleared, with little or no renal clearance, istradefylline exposure may be expected to be higher in patients with hepatic impairment. As CYP3A4/5 is one of the main isoenzymes responsible for istradefylline metabolism, drug-drug interactions with CYP3A inhibitors/inducers may be expected. Systemic metabolite exposure is unlikely to be of clinical relevance since none of the metabolites identified in the mass balance study accounted for >10% of parent drug exposure.

The primary oxidative metabolites of istradefylline were M1 (pharmacologically active, with similar binding affinity for A_{2A} receptors as istradefylline) and M8. Although M1 is an active metabolite, its concentrations in plasma were very low, thus, no significant contribution to the pharmacological activity of istradefylline is expected.

PK in the target population

Study 6002-US-003 was a pilot study to investigate the safety and PK of repeated doses of istradefylline in patients with PD under active treatment with L/C.

On average, steady-state was achieved by 14 days of multiple dosing of istradefylline at 60 and 80 mg/day. The effect of L/C on the PK of istradefylline was not directly evaluated in this study. However, the mean C_{max} and AUC_{0-24} values of istradefylline in subjects with PD under primary treatment with L/C from the current study were comparable to the values obtained for healthy subjects in Study 6002-US-002 who were treated with the same istradefylline doses without L/C.

Systemic exposure to levodopa and carbidopa tended to decrease upon co-administration with multiple doses of istradefylline, suggesting a possible interaction between istradefylline and L/C. However, this study was not designed to investigate the effect of istradefylline on the PK of L/C because each subject was on a different dose of L/C and there were only 5 subjects in each istradefylline dose group.

Special populations

- ***Impaired renal function***

Study 6002-US-015 was a dedicated renal impairment study in non-smoking subjects with severe renal impairment (CrCL<30 mL/min) and reported that the mean istradefylline $AUC_{0-\infty}$ and C_{max} were 16% and 21% lower in subjects with severe renal impairment compared to matched healthy subjects. There was no correlation between important PK parameters and creatinine clearance, and there was no apparent difference in protein binding between renally impaired patients and matched healthy subjects. In terms of the main metabolites M1 and M8, the differences observed between the subjects with renal impairment and the 2 control groups are considered unlikely to be of clinical relevance since both metabolites are present in trace amounts.

The results suggest that impact of severe renal impairment on istradefylline PK is unlikely to be clinically relevant. Istradefylline was not studied in patients with end-stage renal disease and end-stage renal disease requiring dialysis.

- ***Impaired hepatic function***

Study (6002-016) was conducted to evaluate the effect of mild hepatic impairment on single dose PK of istradefylline and its metabolites in non-smoking subjects. The PK exposure of istradefylline was minimally affected by mild hepatic impairment (30% lower C_{max} and 9% lower $AUC_{0-\infty}$). The effect of mild hepatic impairment was variable on the exposure of metabolites; no effect on istradefylline M1 metabolite and 27% lower exposure to istradefylline M8 metabolite. Given the low abundance of metabolites in the plasma (<10%), these differences are not of clinical relevance.

Study 6002-US-016 was conducted to assess the effects of moderate hepatic impairment with and without the added condition of smoking 20 or more cigarettes a day. In both smokers and nonsmokers, the $t_{1/2}$ of istradefylline in subjects with moderate hepatic impairment was significantly prolonged compared to matched controls (by 1.79- and 2.44-fold in smokers and non-smokers, respectively).

In the first (original) analysis of data, steady-state systemic exposure in nonsmoking subjects with moderate hepatic impairment was estimated to be about 3.3 times the value in healthy nonsmoking subjects. However, the methods used were clearly flawed and would likely have resulted in an overestimation of accumulation and hence steady-state istradefylline exposure.

Subsequently, the applicant reanalysed the data using compartmental modelling and simulation. The results of this reanalysis predicted that steady-state exposure would be increased, on average, by around 40% (AUC_{0-24} ratio 1.39; 90% CI 0.93, 2.19) in non-smoking patients with moderate hepatic impairment compared to healthy nonsmokers. Of note, the upper limit of the 90% CI around the mean ratio was

220%, meaning that some patients with moderate hepatic impairment may exceed the safety exposure threshold of 60 mg QD. Therefore, the maximum recommended dose of istradefylline was reduced to 20 mg/day in non-smoking patients with moderate hepatic impairment.

- ***Gender***

Study 6002-EU03 investigated the PK of istradefylline in healthy male and female elderly subjects following single ascending doses. C_{max} and t_{max} were generally similar between healthy older male and female participants following administration of single ascending doses of istradefylline. However, istradefylline AUC values were approximately 1.37 to 1.66-fold greater in females following administration of 50 to 150 mg. However, there was no adjustment for covariates in this analysis.

Subsequent analysis of AUC by dose, smoking status and gender showed that once smoking was accounted for, the PK parameters were similar between genders.

Study PP15710 and Study 6002-US-024 investigated the PK of istradefylline in healthy male and female subjects following multiple dosing for 14 days. There was no evidence of sex-related differences in systemic exposure to istradefylline in either study.

Population PK analysis [6002-014-pop-pk-r-en (2017)]: Based on the final population PK model, the predicted difference in istradefylline exposure between male and female patients is relatively small (14%) and may be driven by differences in body weight.

Population PK analysis 6002-nda-response-pop-pk-r-en (2018): Based on simulations using the final population PK model, the predicted difference in istradefylline exposure between females and males was 32%. The reason for this difference is unclear but it could be due to differences in body weight.

- ***Race***

Pop-PK analysis 6002-nda-response-pop-pk-r-en (2018): Based on simulations using the final population PK model, the predicted difference in istradefylline exposure between Asian and non-Asian patients was 35%. This difference may be driven by differences in body weight.

- ***Weight***

Pop-PK analysis [6002-014-pop-pk-r-en (2017)]: Based on simulations using the final popPK model, istradefylline exposure [AUC(0-τ)ss] is predicted to be 21-38% higher in patients with increased body size (body weight or BMI) and 3-15% lower in patients with reduced body size compared to a typical patient. This is because the final popPK model suggests CL/F decreases with increasing BMI, which is not plausible.

Further modelling of the body size covariate on clearance to assess weight provided evidence that this covariate should not be used in an allometric function and that the inverse relationship of body size on clearance provided a better model to describe the data, but with limited impact. Importantly, the analysis followed the *a priori* plan for base model building, inclusion and exclusion of covariates, and applied different approaches to best capture and describe the variability observed in the data, which was the primary objective.

Pop-PK analysis 6002-nda-response-pop-pk-r-en (2018): Based on simulations using the final popPK model, istradefylline exposure [AUC(0-τ)ss] is predicted to be 34% higher in patients with high body weight (102 kg) and 26% lower in patients with low body weight (41 kg) compared to a typical patient (65 kg). This is because the final model suggests CL/F decreases with increasing weight, which is not plausible.

The applicant contends that the final model was statistically valid despite the inclusion of an inverse relationship between clearance and weight. The applicant postulates that there may be an interaction

among covariates that leads to the weight relationship acting as a correction to clearance that complements other covariates. This seems plausible. When the applicant re-evaluated the model with allometric exponents on CL/F and V/F, the relationship between CL/F and body weight was statistically insignificant. This suggests that there is no or a minimal effect of weight on exposure. The applicant also showed that even with the inclusion of weight with a negative effect, there was a strong correlation between exposure parameters derived from the base and final models. Overall, the applicant's response is accepted.

- ***Elderly***

Study 6002-0205 investigated the effect of age on istradefylline PK. No significant difference was found in the PK parameters of istradefylline between healthy elderly and non-elderly male volunteers following administration of a single 40 mg dose. However, the sample size was small (n=9 subjects per group) and, therefore, the study may have been underpowered.

In the pop-PK analysis 6002-pop-pk-analysis (2006), age was identified as a significant covariate on apparent clearance. However, the effect was small and not clinically relevant. Age was not identified as a significant covariate in later pop-PK analyses [6002-014-pop-pk-r-en (2017) and 6002-nda-response-pop-pk-r-en (2018)].

PK data show that the exposure in terms of C_{max}, AUC and C_{min} are similar among groups of age (<65; 65-74; 75-84 and >85 years) and within study, nevertheless a high variability around each PK parameter and a poor representativeness in older group (>85 years).

Interactions

- ***In vitro***

The Summary of Clinical Pharmacology Studies states that the DDI risk assessment using a basic model predicted that istradefylline has minimal DDI risk of *in vivo* inhibition of BCRP, OATP1B1, OATP1B3, OAT1, OCT2, MATE1, and MATE2-K (r-18-0051), which is not accurate. In study r-18-0051, the ratio of gut luminal concentration to inhibition potencies (Igut/IC50) of istradefylline for BCRP was 6090, which greatly exceeds the cut-off value of 10, suggesting that istradefylline has the potential to inhibit BCRP *in vivo*. The applicant contends that the risk for a clinically meaningful *in vivo* DDI when istradefylline is co-administered with another drug that is a BCRP substrate such as methotrexate is low.

- ***In vivo***

Study BP15809 investigated the effect of istradefylline on the PK of the CYP3A substrate midazolam. The results showed that treatment with istradefylline at once daily doses of 5 or 20 mg for 14 days had a minimal effect on the mean AUC_{0-24} of midazolam, suggesting that istradefylline at these doses did not affect CYP3A4 activity due to inhibition and/or induction. However, the systemic exposure of the potential perpetrator drug should be the exposure obtained with the highest generally recommended dose under therapeutic (steady state) conditions. For istradefylline, the highest recommended dose is 40 mg/day. Therefore, the istradefylline doses evaluated in this study are considered too low to conclude a lack of CYP3A inhibition.

Study 6002-US-008 investigated the *in vivo* interaction potential of istradefylline with CYP3A4.

Midazolam interaction

The applicant conducted an *in vivo* drug interaction study (6002-US-008) to evaluate the impact of istradefylline as a potential CYP3A4 inhibitor using midazolam as a model substrate. In this cohort 10 mg of midazolam was administered either (a) alone or (b) co-administered with istradefylline (80 mg/day for 15 days).

Istradefylline significantly increased the peak exposure (C_{max}) and total exposure ($AUC_{0-\infty}$) of midazolam by 1.6-fold and 2.4 fold, respectively, suggesting that istradefylline is a moderate inhibitor of CYP3A4 at a dose of 80 mg/day. The increase in systemic exposure of the parent compound, which was not accompanied by a significant decrease in the systemic exposure of the metabolite, suggests a predominant pre-systemic effect of istradefylline on CYP3A4 activity.

The dose of 80 mg istradefylline evaluated in this study is 2-4-fold higher than the recommended dose. No significant effects were observed at the 20 mg dose in Study BP15809. A PBPK model was constructed to understand the effect of 40 mg istradefylline on the PK of midazolam. Considering verification and qualification results, the PBPK model was deemed fit for purpose. Based on the model, a weak interaction (AUC ratio ≥ 1.25 and < 2) was predicted between 40 mg istradefylline and midazolam. This weak inhibitory effect caused by istradefylline may not have clinical relevance for the victim drug unless the victim drug has narrow therapeutic index.

Ketoconazole interaction

In the same study (6002-US-008), in another cohort, the applicant evaluated the impact of ketoconazole (a strong CYP3A inhibitor) on istradefylline. The PK of istradefylline was compared following a single dose of 40 mg istradefylline administered alone vs when co-administered with ketoconazole (200 mg twice daily for 4 days). The dosing regimen of ketoconazole ensured complete inhibition of CYP3A4 before istradefylline treatment was commenced and continued inhibition of CYP3A4 activity for an additional 7 days after this.

Statistical evaluation of PK data indicated that the extent of istradefylline exposure as measured by AUC_{0-72} and AUC_{0-168} on Day 19 was modestly increased by 1.3- and 1.5-fold, respectively, when compared to Day 1. The increase in $AUC_{0-\infty}$ was 2.5-fold and the elimination half-life increased by 187 hours. However, the plasma PK was only collected for 168 hours postdose, and several subjects had extrapolated areas that comprised greater than 60% of the $AUC_{0-\infty}$, suggesting the possibility of overestimation of $AUC_{0-\infty}$. Simulations based on a PBPK model predicted an increase in AUC_{inf} of ~2-fold.

In the clinical development program, istradefylline was studied in doses up to 60 mg QD, thus establishing the safety profile into this exposure range. Therefore, for strong CYP3A inhibitors, like ketoconazole, dosing is limited to 20 mg QD to stay within this range.

No studies have been conducted to evaluate the effect of concomitant use of a mild and moderate CYP3A inhibitors on istradefylline PK. Instead, a PBPK model was constructed to evaluate the effects of mild and moderate CYP3A4 inhibitors on istradefylline PK. Considering verification and qualification results, the use of PBPK data to predict these interactions was deemed acceptable. The simulations predicted no interaction between mild inhibitors and istradefylline, and a potential weak interaction between moderate inhibitors and istradefylline.

Study 6002-US-020 evaluated the effect of istradefylline as a potential CYP3A inhibitor using atorvastatin as a substrate. In this study, 40 mg of atorvastatin was administered either (a) alone or (b) co-administered with istradefylline (40 mg/day for 17 days). These doses are considered appropriate to investigate this drug interaction.

The results showed that istradefylline increased atorvastatin C_{max} by 53% and $AUC_{0-\infty}$ by 54% compared with atorvastatin alone. The t_{max} of atorvastatin was significantly shorter and the mean $t_{1/2}$ of atorvastatin increased by 27% (2.5 hours) in the presence of istradefylline. This was not accompanied by any significant changes in the exposure of either of its 2 metabolites, which suggests that the effect of istradefylline on atorvastatin PK is primarily presystemic.

Study 6002-015 examined the DDI between rifampicin, a strong CYP3A4 inducer, and istradefylline in 20 healthy subjects. The dosage regimens used in the study are appropriate. Istradefylline 40 mg is the

highest proposed dose. A 7-day administration of rifampicin, generally, reaches maximal induction of CYP3A4. Rifampicin dosing with 600 mg daily was continued for 20 days which is adequate to ensure continued CYP3A induction over the istradefylline PK sampling period of 4 to 5 $t_{1/2}$ of istradefylline.

When coadministered with rifampicin, there was a considerable decrease in the exposure of istradefylline. The geometric mean ratio of C_{max} and $AUC_{0-\infty}$ were 55.50% (90% CI: 49.45% to 62.29%) and 19.16% (90% CI: 17.87% to 20.54%), respectively, with coadministration of rifampicin. This means that C_{max} and $AUC_{0-\infty}$ were reduced by 44.5% and 80.8% when coadministered with rifampicin, respectively. The effects of rifampicin on the istradefylline M1 and M8 metabolites were inconsistent and variable. However, the rifampicin effect on the 2 metabolites is not considered clinically relevant as the metabolite/parent ratios for AUC_{0-last} or $AUC_{0-\infty}$ for M1 and M8 were less than 10%.

The decrease of istradefylline exposure by 80% due to concomitant administration of a strong CYP3A4 inducer is considered clinically significant.

No clinical study has been conducted to investigate the impact of moderate CYP3A4 inducers on istradefylline PK. Instead, a PBPK model was built to understand the effect of the moderate CYP3A inducer efavirenz on istradefylline PK. However, model verification showed an over-prediction of simulated profiles of istradefylline in the presence of rifampicin and the qualification dataset was very limited. Further, supportive data currently available from public literature are not considered sufficient for qualification of SIMCYP platform for CYP3A mediated induction (unlike inhibition). Therefore, the SmPC states that use of istradefylline with moderate CYP3A4 inducers is not recommended.

Study 6002-US-026 evaluated the effect of istradefylline as a potential P-gp inhibitor using digoxin as a P-gp substrate. In this study, 0.4 mg of digoxin was administered either (a) alone or (b) co-administered with istradefylline (40 mg/day for 21 days). These doses are considered appropriate to investigate this drug interaction.

The results showed that there was a mild drug interaction between steady-state istradefylline and digoxin. Istradefylline increased the peak exposure (C_{max}) of digoxin by 33% and total exposure ($AUC_{0-\infty}$) by 21% compared to digoxin administration alone. The $t_{1/2}$ of digoxin was not changed in the presence of istradefylline.

The magnitude of this DDI is considered small and may not be clinically significant. However, caution should be exercised for narrow therapeutic index drug such as digoxin.

Study BP15748 was conducted to investigate the potential PK interaction of concomitant administration of repeated doses of istradefylline together with repeated doses of L/C. This is important because istradefylline is indicated as an adjunctive treatment to L/C in patients with PD.

The results indicated that the steady-state PK of L/C were not significantly affected by co-administration of repeated doses of istradefylline. It is noted that ratios of LSM test/reference were presented with 95% CI, rather than 90% CI which would have been narrower.

A comparison of istradefylline PK parameters derived in the current study with those from study PP15710 (a multiple ascending dose study in elderly subjects) suggested that there is no significant effect of L/C on the PK of istradefylline. However, the comparison is not considered robust since it was not directly assessed in a single clinical study. This issue was not pursued further because istradefylline used concomitantly with L/C was evaluated in all pivotal Phase 3 clinical studies.

Study 6002-US-009 evaluated the effects of 14 days of oral administration of 80 mg istradefylline on the PK of a single dose of L/C. The results demonstrated that istradefylline does not affect the PK of L/C, which is consistent with the findings of Study BP15748.

Study PP15710 evaluated the effect of smoking on istradefylline PK. The apparent clearance of istradefylline was around 3-fold higher in smokers compared to nonsmokers and, therefore, systemic exposure was significantly lower. The mean ratios for C_{max} and AUC_{0-24} (smokers vs. non-smokers) on Day 14 were 0.56 and 0.37, respectively. This suggests that higher doses of istradefylline may be needed in patients that smoke.

Study 6002-US-016 evaluated the impact of smoking (> 20 cigarettes/day) in healthy patients and patients with moderate hepatic impairment. Consistent with the findings of Study PP15710, the PK parameters of istradefylline were significantly altered in smoking subjects. In both hepatically impaired and matched-control subjects, smokers had significantly lower exposure compared to nonsmokers. The LSM ratio for AUC values were around 20-30% in smokers compared to nonsmokers. Moreover, the mean $t_{1/2}$ was shortened from 288 hours in hepatically impaired nonsmokers to approximately 100 hours in hepatically impaired smokers, and from 118 hours in healthy nonsmokers to approximately 56 hours in healthy smokers.

Based on these results, the recommended istradefylline dosage in patients who smoke 20 or more cigarettes per day (or the equivalent amount of another tobacco product) is 40 mg once daily.

Population PK-PD analysis [6002-POP-PK-ANALYSIS (2006)] conducted simulations to determine dose recommendations for istradefylline in patients who smoke. According to simulations, an increase in dose from 20 to 40 mg/day for the smoker would result in a predicted response that would be similar to the response demonstrated in a non-smoker who receives 20 mg/day. For an istradefylline response equivalent to the effect expected at 40 mg/day in non-smokers, smokers would require a dose of approximately 60 mg/day.

Pharmacodynamics

Mechanism of action

Istradefylline is a selective adenosine A_{2A} receptor antagonist. It is hypothesized that blockage of the A_{2A} receptors by istradefylline reduces the excitability of the striatal pathway that occurs in PD, resulting in an improvement in PD symptoms.

Primary pharmacology

In Study 6002-EU06, PET showed that over 90% adenosine A_{2A} RO was achieved with daily oral doses of greater than 5 mg istradefylline for 14 days. A dose-occupancy curve could be constructed following 0.1 to 5 mg daily for 14 days. Dosing with 40 mg, 20 mg and 5 mg daily for 14 days achieved almost complete suppression of the binding of radiolabelled istradefylline.

The results do not appear to support the proposed minimum dose of 20 mg/day, since the 5 mg dose achieved similar RO and, thus, should have similar effectiveness. However, this study was conducted in healthy subjects and there is a growing body of evidence that in the brains of patients with PD, adenosine A_{2A} receptor density is greater compared with healthy subjects. Thus, the positron emission tomography scan results from healthy subjects are not necessarily predictive for the selection of the minimally effective dose of istradefylline in patients with PD.

Secondary pharmacology

Study 6002-US-024 was a thorough QTc study to examine the effect of istradefylline on ECG parameters. The study design was appropriate and the sample size was adequate. The duration of treatment of 14 days was adequate to achieve steady state istradefylline concentrations. The plasma exposures achieved in this study are considered adequate to cover the range that could be expected in PD at recommended doses of 20-40 mg/day. The ketoconazole interaction study demonstrated a 2.5-fold (90%CI 1.9-3.2-fold) increase in the $AUC_{0-\infty}$ of istradefylline. Exposure at a supratherapeutic dose of 160 mg/day

istradefylline used in this study would exceed the increase in exposure caused by ketoconazole. Assay sensitivity at the 5 millisecond level was demonstrated with the active control moxifloxacin.

The results of the study showed that istradefylline does not affect AV conduction, depolarisation, or cardiac repolarisation as measured by the PR, QRS, or QTcI interval durations to a clinically important extent. There was no effect on heart rate except that at the istradefylline clinical dose of 40 mg daily at steady state more subjects (40%) as compared with placebo (15%) met the tachycardic outlier criteria. However, this is considered unlikely to be a drug-related finding since the effect was similar for 160 mg istradefylline (21%) and placebo (15%).

The LSM time-averaged mean QTcI change from baseline (placebo corrected) for istradefylline 40 mg was -1.7 millisecond on both Day 1 and Day 14. Istradefylline 160 mg demonstrated a placebo corrected mean change of -0.3 millisecond on Day 1 and 0.2 millisecond on Day 14. For the time-matched analysis, the upper limit of the 90% CI for placebo corrected change from Baseline for istradefylline 160 mg at steady state slightly exceeded 10 milliseconds by up to 0.4 milliseconds at 3 time points, but the 40 mg dose had no increases above 10 milliseconds at all timepoints. This could be due to the tachycardic outliers in the 160 mg dose group.

Overall, the results do not raise any particular concerns in terms of ECG parameters.

In Study 6002-017, the abuse potential of istradefylline (40 mg, 80 mg, and 160 mg) was evaluated using the stimulant phentermine as an active comparator in a highly sensitive population of recreational stimulant users. The overall design, including the subject population, selection of doses and positive control were appropriate for the assessment of abuse potential of istradefylline. The study was adequately powered. Study validity was confirmed by the statistically significant difference between phentermine and placebo on the primary endpoint of Drug Liking VAS E_{max}. The mean peak istradefylline concentrations observed in study subjects were generally comparable to those observed in other single dose clinical studies in healthy subjects at similar doses.

Istradefylline doses of 40 mg, 80 mg, and 160 mg showed statistically greater effects than placebo on the primary endpoint of Drug Liking VAS E_{max} and some secondary endpoints of balance of effects, positive effects, stimulant effects and any effects. However, the differences were smaller than those observed with phentermine (i.e., mean differences on the primary endpoints ranging from ~4 to 5 points on a 100-point scale vs. ~ 14 and 21 points with phentermine 45 and 90 mg).

The applicant contends that the mean differences for istradefylline are not consistent with clinically important abuse potential because they are less than the minimum ~15-20 point differences from placebo that are observed with drugs of abuse across multiple drug classes (Schoedel et al, 2012b). Further, the upper limit of the 95% CIs of differences in Drug Liking VAS E_{max} between all 3 istradefylline doses and placebo were <11 points; a proposed equivalence margin for determining clinically meaningful differences from placebo, based on a meta-analysis of human abuse potential studies (Chen & Bonson, 2013). However, no equivalence margin was pre-specified and justified in the study protocol.

The applicant further evaluated istradefylline's potential for abuse and its potential liability to produce tolerance and/or physical dependence upon withdrawal. This comprehensive evaluation concluded that istradefylline has a low risk of abuse potential based on the evidence from nonclinical sources, clinical studies (including Study 6002-017, a human abuse liability study in recreational stimulant users) and post-marketing experience. The applicant's conclusion is supported.

Relationship between plasma concentration and effect

6002-pop-pk-analysis (2006): Population PK-PD analysis of istradefylline in healthy subjects and in patients with PD

The PK-PD models developed to assess the relationship between istradefylline exposure and efficacy endpoints were able to describe the observed data reasonably well. The results suggest a relatively small effect of istradefylline on efficacy endpoints at the recommended doses, with a shallow dose-exposure-response relationship. At istradefylline doses of 20 mg/day and 40 mg/day, the predicted reduction in actual OFF time is 0.64 hr (38 mins) and 0.75 hr (45 mins), respectively. Doses greater than 40 mg/day are predicted to yield minimal increases in efficacy.

The PK-PD models developed to assess the relationship between istradefylline exposure and adverse events of dyskinesia, dizziness and nausea did not, in general, describe the observed data well. In each of the 3 PK-PD models developed, the model parameters tended to be poorly estimated, with some exceptions. The results suggest that istradefylline doses greater than 40 mg/day would yield minimal to no increases in the incidence of dizziness and probability of dyskinesia, but could result in an increased incidence of nausea AE.

Covariate effects included in the final models tended to be poorly estimated and in the majority of cases the bootstrap 95% CIs for the estimate included the null value. Therefore, the impact of covariates on efficacy and safety endpoints remains unclear.

6002-014-pkpd-r-en (2017): Exposure-response analysis of istradefylline

In this E-R analysis, none of the efficacy endpoints tested demonstrated an E-R relationship with istradefylline. In fact, istradefylline at 20 mg/day and 40 mg/day, when added as adjunctive therapy, did not show a statistically significant difference from placebo for any of the efficacy endpoints.

The overall adverse event rate was low. Only the incidence of nausea, dyskinesia and falls exceeded 5%. Of these, dyskinesia exhibited increased frequency with increasing exposure and a significantly higher odds ratio at higher levels of exposure. The frequency of nausea also increased with increasing exposure with a significantly higher odds ratio at higher levels of exposure.

6002-nda-response-exposure-response-analysis-r-en (2018): Exposure-response analysis of istradefylline (2018)

In this analysis, PK data and clinical response data for the primary endpoint (OFF time reduction) and for the frequency of selected AEs from 5 studies (6002-US-005, 6002-US-006, 6002-US-013, 6002-0608, and 6002-009) were analysed to explore the E-R relationships (efficacy and safety) for istradefylline.

Consistent with the findings of the E-R analysis 6002-014-pkpd-r-en (2017), there was no apparent exposure-response relationship for the efficacy endpoint OFF time change from baseline across the 20-60 mg/day dose range. However, OFF time change from baseline in active istradefylline treatment groups was significantly different from placebo at these doses, even though the actual reduction in OFF time with istradefylline treatment compared to placebo was small.

There was no apparent relationship between incidence of AEs and exposure to istradefylline except for an increase in liver enzyme elevations with increasing exposure. However, there were no serious liver elevations and the frequency of liver enzyme elevations in the highest AUC quartile was 4.7%. Overall, the results of the exposure-response analyses provide limited support for a dose titration strategy for istradefylline from 20 mg/day to 40 mg/day. The applicant's rationale for having this dose range is that, in clinical practice, the management of motor fluctuations or dyskinesia is individualized and strongly based on the patient's preference and tolerability. Further, data in the 8 pivotal studies show that greater benefit may be achieved with the 40 mg/day dose compared with the 20 mg/day dose, and additional benefit was observed in some patients who had their dose increased from 20 mg/day to 40 mg/day in Study 6002-010. In addition, PK/PD data suggest that a higher dose of 40 mg/day may be more effective

in some patients, since the EC_{50} was exceeded with the 20 mg/day dose but the EC_{75} was only exceeded by 40 mg/day dose

2.4.5. Conclusions on clinical pharmacology

The applicant presented a large number of clinical pharmacology studies for this application. No major objections were raised in terms of the clinical pharmacology. All other concerns have been sufficiently resolved.

2.5. Clinical efficacy

The applicant has submitted 8 pivotal Phase 2b/3 randomized, DB, fixed-dose, placebo controlled studies and 2 supportive long-term, OL studies as clinical evidence of istradefylline for the target indication, 'as an adjunctive treatment to levodopa-based regimens in adult patients with PD experiencing "OFF" time.'

2.5.1. Main studies

Table 35: The following were the 8 pivotal clinical trials.

6002-US-005	6002-US-006	6002-US-013	6002-0608	6002-009	6002-US-018	6002-EU-007	6002-014
Design: Double-blind, randomized, placebo-controlled, parallel-group clinical study							
Duration:							
12-week	12-week	12-week	12-week	12-week	12-week	16-week	12-week
Treatment Groups (randomization ratio):							
Istradefylline 40 mg/day or placebo (2:1 ratio)	Istradefylline 20 or 60 mg/day or placebo (2:2:1 ratio)	Istradefylline 20 mg/day or placebo (1:1 ratio)	Istradefylline 20 or 40 mg/day or placebo (1:1:1 ratio)	Istradefylline 20 or 40 mg/day or placebo (1:1:1 ratio)	Istradefylline 10, 20, or 40 mg/day or placebo (1:1:1:1 ratio)	Istradefylline 40 mg/day, or placebo, or entacapone (1:1:1 ratio)	Istradefylline 20 or 40 mg/day or placebo (1:1:1 ratio)

Hauser Diary:

The primary efficacy variable and some secondary variables of all pivotal studies were based on the subject's diary records.

Subjects were to record the ON/OFF-times in their diary charts on days preceding each visit. Subjects were trained to identify whether the majority of a 30-minute period was spent as either ON or OFF (e.g. 20 minutes at OFF and 10 minutes at ON was to be rated as OFF).

For each 30-minute period during the day, subjects (with the help of a member of the family or other caregiver) were to rate their mobility as:

- OFF: poor mobility or complete immobility (You can move only slowly or not at all).
- ON with troublesome dyskinesia: limited mobility (You are able to move around despite presence of troublesome dyskinesia).
- ON with non-troublesome dyskinesia: good mobility (You are able to move around relatively well despite presence of dyskinesia).
- ON without dyskinesia: excellent mobility (You are able to move around well).
- Asleep

The selection of the PD diary for this study was based on the home diary, developed by Hauser et al., which is considered a reliable and well-validated instrument that assesses functional status in Parkinson's disease patients with motor fluctuations and dyskinesias. The diary is constructed so that on specified days, the patient records their status for each 30-minute interval as 1 of 5 categories: Asleep, OFF time, ON time without dyskinesia, ON time with non-troublesome dyskinesia, or ON time with troublesome dyskinesia. This instrument allows characterization of patient status as "bad" time (i.e., OFF time plus

ON time with troublesome dyskinesia) or "good" ON time (i.e., ON time without troublesome dyskinesia defined as ON time without dyskinesia plus ON time with non-troublesome dyskinesia).

Definitions

The following definitions were used in all pivotal trials:

- ON time: the time when medication is providing benefit with regard to mobility, slowness, and stiffness
- OFF time: the time when medication had worn off and is no longer providing benefit with regard to mobility, slowness, and stiffness.
- Dyskinesia: involuntary twisting, turning movements (tremor, i.e., shaking back and forth, was not considered dyskinesia)
- Non-troublesome dyskinesia: dyskinesia that did not interfere function or cause meaningful discomfort.
- Troublesome dyskinesia: dyskinesia that interfered with function or caused meaningful discomfort.

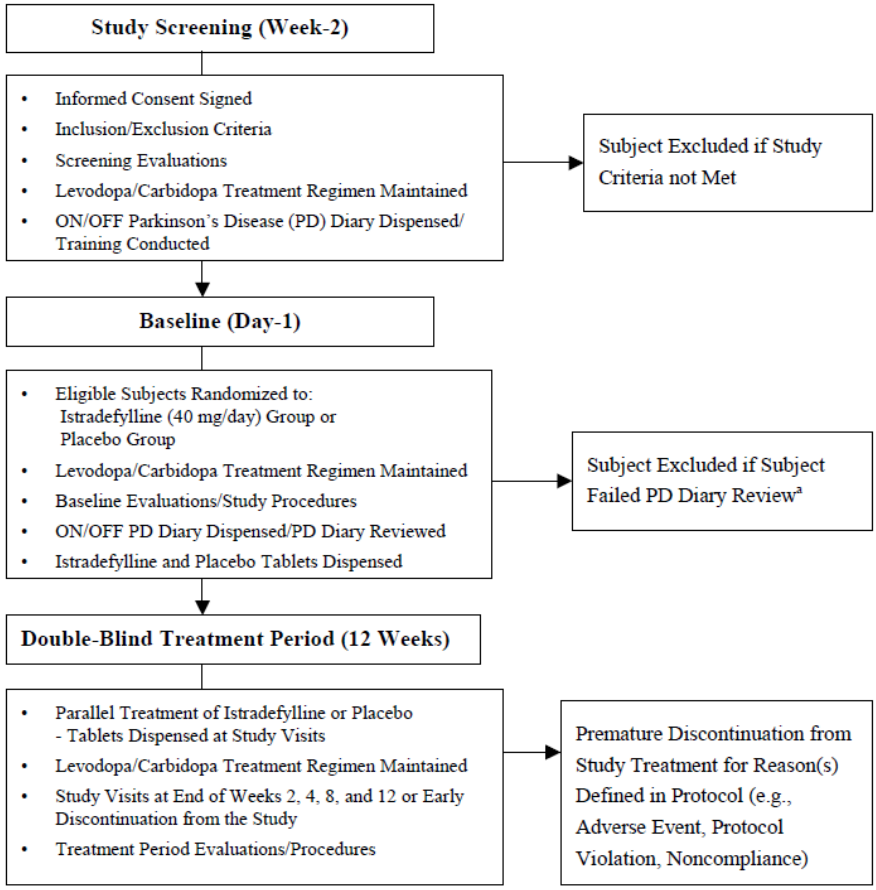
Study 6002-US-005

"A 12-Week, DB, Placebo-Controlled, Randomized, Multicenter Study of the Efficacy of 40 mg/day KW-6002 as Treatment for PD in Patients with Motor Response Complications on Levodopa/Carbidopa Therapy"

Methods

Study design

Figure 24: Study Plan of Study 6002-US-005



All subjects underwent Screening evaluations over a period of at least 2 weeks. Subjects were to maintain their regular L/C regimen during screening and DB treatment according to the Investigator's clinical judgment and the requirements of the protocol. During Screening (Week -2), subjects were to be instructed on the proper completion of the ON/OFF PD diary and were to complete two ON/OFF PD diaries for training purposes; at least an 80% concordance with subject's and Investigator's ratings of the subject's ON and OFF time (excluding dyskinesia) was to be established.

At the Baseline visit (Day -1), eligible subjects were randomized to treatment with 40 mg/day istradefylline or matching placebo in a 2:1 ratio. Subjects were to return at Weeks 2, 4, 8, and 12 for assessment of symptoms of PD and safety evaluations. Subjects were to complete the ON/OFF PD diaries on any 2 consecutive days during the 7-day period preceding the Baseline visit (Day -1) and each subsequent scheduled visit (Weeks 2, 4, 8, and 12).

In order to evaluate motor symptoms in an OFF state, subjects were instructed not to take study drug or any other antiparkinson medications after midnight on the day prior to 3 scheduled visits (Day -1, Week 4, and Week 12). The UPDRS subscale III assessments were performed prior to dosing (i.e., the subject was likely to be in an OFF state) at these visits. After completion of the UPDRS subscale III assessments, study drug and the usual dose of other antiparkinson medications were administered to the subject (i.e., the subject was likely to be in an ON state), and the UPDRS subscale III assessments were repeated.

Study participants

Inclusion Criteria

- Met the United Kingdom Parkinson's Disease Society (UKPDS) brain bank diagnostic criteria (step 1 and 2) for PD.
- PD in Stages 2-4 while in an OFF state on the modified Hoehn and Yahr Scale (H&Y).
- Had responded to L/C, had been treated with L/C for at least 1 year, and had been on a stable regimen of L/C for at least 4 weeks before randomization.
- Were currently taking at least 4 doses of L/C per day (three doses per day if at least 2 doses contained slow-release formulation) and with predictable end-of-dose wearing off.
- Had successfully completed PD diary training at the Week -2 visit and completed 2 diaries on any 2 consecutive days during the week (7-day period) before the Day –1 visit, excluding the day immediately prior to that visit. The subject must have had an average of at least 120 minutes of OFF time on the 2 diaries and not more than 4 invalid entries were allowed on either of the mandatory training diaries.
- Have been on a stable regimen of medications being administered within normal therapeutic limits for PD for at least 4 weeks before randomization. (Included L/C (including sustained-release and dispersible formulations), centrally acting dopamine agonists (i.e., bromocriptine, pergolide, pramipexole, and ropinirole), and other medications for PD which the subject may have been taking (including anticholinergics, selegiline, antihistamines and β-blockers)).
- At least 30 years of age.
- Able to give written informed consent.

Exclusion criteria

- Subjects treated with liquid L/C within the 4 weeks before randomization;
- Subjects who were currently being treated with any excluded medications as established in the protocol.
- Subjects who were treated within 30 days (or 5 half-lives of the compound, if longer) before randomization with any investigational agent.
- Subjects taking monoamine oxidase inhibitors, except selegiline.
- Subjects treated within 3 months before randomization with centrally acting dopamine antagonists (6 months if subject was treated with depot formulation); e.g., antipsychotic neuroleptics, metoclopramide, buspirone, amoxapine. Exceptions to the exclusion of subjects who were taking antipsychotic medications must have been determined on an individual case basis in discussion with the medical monitor.
- Neurosurgical operation for PD (e.g., pallidotomy, thalamotomy, deep brain stimulation).
- Subjects previously treated with istradefylline.
- Subjects with atypical parkinsonism or secondary parkinsonism variants.
- Subjects with a diagnosis of cancer or evidence of continued disease within 5 years of study enrollment (except for subjects who had had basal cell carcinoma or carcinoma in situ of the cervix surgically excised), or a clinically significant illness of any organ system, including the hepatic (ALT and/or AST value greater than 1.5 times the upper limit of normal [ULN]), renal, pancreatic, cardiovascular, endocrinologic, gastrointestinal, respiratory, and neurologic systems (except PD) who may have required a change in the treatment of that illness during the study, or which may have compromised the safety of the subject during the study or affected the ability of the subject to complete the study;
- Subjects who, for any reason, were judged by the Investigator to be inappropriate for this study, including subjects who were unable to communicate or to cooperate with the Investigator.
- Subjects with significant dementia or with Mini-Mental State Examination (MMSE) score of 25 or less.
- Subjects with a history of drug or alcohol abuse or dependence (Disease State Management-IV criteria) within the past 2 years.
- Subjects with a history of a psychotic illness, unless the episodes were brief or were drug

induced.

- Subjects with current clinically relevant depression disorder.
- Subjects with a history of seizures, with the exception of a single febrile seizure during infancy or childhood.
- Subjects with a history of neuroleptic malignant syndrome.
- Pregnant or lactating females. Females of childbearing potential must have had a negative result on their serum pregnancy test performed on Day -1. Sexually active females of childbearing potential were to have used a reliable method of contraception.

Treatments

The study treatments were 40 mg istradefylline or matching placebo. All istradefylline and placebo tablets were supplied by KPI.

The daily dose of istradefylline or placebo (1 tablet) was to be taken in the morning with food(breakfast) for 12 weeks. Study drug could be taken without food if the subject did not routinely eat breakfast.

During the study, subjects were to take L/C according to the Investigator's clinical judgment and the requirements of the protocol. In addition, other antiparkinson medications were permitted L/C and any other antiparkinson medications were to be obtained from local pharmacies.

Levodopa Adjustment

After randomization, no changes were to be made to the levodopa treatment regimen without first consulting the Investigator. Decreases in the total daily dose of L/C because of AEs were permitted. After the AEs had been alleviated, increases in the total daily dose of L/C to the baseline level were permitted. Changes in the interval between L/C doses were not allowed.

Adjustment of Other Antiparkinson's Medications

After the subject was randomized, doses of other antiparkinson medications were to be decreased to control levodopa-related adverse events only after the following:

- an attempt was made to adjust the subject's L/C regimen and this was unsuccessful, or
- it was determined that the AE was specifically attributed to that agent (e.g., urinary retention because of an anticholinergic).

Subjects were not permitted to make any changes in their treatment regimens without first consulting the Investigator. The addition of new antiparkinson medications or an increase in dose of antiparkinson medications were not permitted during the study. In the case of an adverse event because of a specific agent, the dose of antiparkinson medication could have been decreased or the medication could have been stopped. All changes in antiparkinson medication (either dosage or regimen) were to be recorded in the source documents and on the case report form (CRF).

The use of medications, other than study drug and other antiparkinson agents, was limited to treatments for pre-existing medical conditions or adverse events. Subjects who took domperidone or supplemental carbidopa were to keep the dose constant throughout the study (from Week -2 until the study completion, or early discontinuation).

Treatment duration and response assessment

The daily dose of istradefylline or placebo (1 tablet) was to be taken in the morning with food (breakfast) for 12 weeks. Subjects were to return at Weeks 2, 4, 8, and 12 for assessment of symptoms of PD and safety evaluations. Subjects were to complete the ON/OFF PD diaries on any 2 consecutive days during

the 7-day period preceding the Baseline visit (Day -1) and each subsequent scheduled visit (Weeks 2, 4, 8, and 12).

In order to evaluate motor symptoms in an OFF state, subjects were instructed not to take study drug or any other antiparkinson medications after midnight on the day prior to 3 scheduled visits (Day -1, Week 4, and Week 12). The UPDRS subscale III assessments were performed prior to dosing (i.e., the subject was likely to be in an OFF state) at these visits. After completion of the UPDRS subscale III assessments, study drug and the usual dose of other antiparkinson medications were administered to the subject (i.e., the subject was likely to be in an ON state), and the UPDRS subscale III assessments were repeated.

Objectives

The primary objective of this study was to evaluate the safety and establish the efficacy of 40 mg/day istradefylline for reducing OFF time in subjects with advanced PD treated with L/C.

The secondary objective of this study was to establish the efficacy of 40 mg/day istradefylline for reducing motor symptoms and improving activities of daily living (ADL) in subjects with advanced PD treated with L/C.

Outcomes/endpoints

Primary efficacy endpoint

The primary efficacy variable of this study was the change from baseline in percentage of awake time per day spent in an OFF state at Endpoint based on the subjects' valid ON/OFF PD diary data. The endpoint was defined as the Week 12 value or the last available post-baseline value at the time of the subject's discontinuation from the study. To meet this objective, subjects recorded assessments of ON and OFF time every 30 minutes for 24 hours in the subjects' ON/OFF PD diary. See definitions above.

Secondary efficacy measurements were based on the changes in the subjects' valid ON/OFF PD diary data, the UPDRS scale, and the Clinical Global Impression – Improvement scale (CGI-I scale). The definition of ON and OFF times for the secondary efficacy variables was the same as that provided for the primary efficacy variable.

Secondary efficacy endpoints

1. Based on ON/ OFF PD Diary Data (validated for completion by subjects)

Actual values and change from baseline in the percentage or total hours of awake time per day spent in the defined state were displayed as follows:

OFF State:

- Percentage of awake time per day spent in an OFF state at Weeks 2, 4, 8, and 12 (including actual values at Endpoint).
- Total hours of awake time per day spent in an OFF state at Weeks 2, 4, 8, 12, and Endpoint.

ON State Without Dyskinesia:

- Percentage of awake time per day spent in an ON state without dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.
- Total hours of awake time per day spent in an ON state without dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.

ON State With Dyskinesia:

- Percentage of awake time per day spent in an ON state with dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.
- Total hours of awake time per day spent in an ON state with dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.

ON State With Non-Troublesome Dyskinesia:

- Percentage of awake time per day spent in an ON state with non-troublesome dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.
- Total hours of awake time per day spent in an ON state with non-troublesome dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.

ON State With Troublesome Dyskinesia:

- Percentage of awake time per day spent in an ON state with troublesome dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.
- Total hours of awake time per day spent in an ON state with troublesome dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.

Analyses of total hours of awake time spent in an ON state with non-troublesome dyskinesia and with troublesome dyskinesia were added after the unblinding of treatment codes.

The following displays for ON state without troublesome dyskinesia (i.e., the sum of ON state without dyskinesia and ON state with non-troublesome dyskinesia) were added after the unblinding of treatment codes:

ON State Without Troublesome Dyskinesia:

- Percentage of awake time per day spent in an ON state without troublesome dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.
- Total hours of awake time per day spent in an ON state without troublesome dyskinesia at Weeks 2, 4, 8, 12, and Endpoint.

2. Based on UPDRS Scores

Subscales I (mentation, behaviour, and mood), II (ADL, assessed in both ON and OFF states), III (motor examination, assessed in both ON and OFF states), IVA (complication of therapy: dyskinesia), IVB (complication of therapy: clinical fluctuation), and IVC (complication of therapy: other complications [anorexia, nausea, vomiting, sleep disturbances, symptomatic orthostasis]) were assessed at Weeks 2, 4, 8, and 12.

Actual values and change from Baseline in UPDRS scores were displayed as follows:

- UPDRS I-IV total score (including ON time rating for UPDRS subscale II and UPDRS subscale III), defined as the sum of subscales I, II, III, and IV at Weeks 2, 4, 8, 12, and Endpoint.
- UPDRS subscale II total score in an ON state at Weeks 2, 4, 8, 12, and Endpoint.
- UPDRS subscale III total score in an ON state at Weeks 2, 4, 8, 12, and Endpoint.
- UPDRS subscale III total score in an OFF state at Weeks 4, 12, and Endpoint.

3. Based on Clinical Global Impression Scores

CGI-I was assessed at Weeks 2, 4, 8, and 12

Clinical Global Impression Scale - Severity of Illness (CGI-S; a 7-point scale, ranging from 1 = normal, not at all ill to 7 = among the most extremely ill subjects) was recorded at baseline only.

Sample size

The sample sizes of the treatment groups were based on the expected difference between treatment groups in the percentage of time OFF from baseline to Endpoint for the intended to treat (ITT) population. A total of 120 subjects in the istradefylline group were planned to be randomized to obtain at least 100 subjects in the ITT population. For the placebo group, 60 subjects were planned to obtain at least 50 subjects in the ITT population. The sample size of each treatment group was sufficient to provide 80% power to detect statistical significance at the 2-sided alpha level of 0.050 for treatment group differences greater than or equal to 50% of the applicable standard deviation. The actual numbers of subjects analyzed in the primary analysis were 129 and 66 subjects, respectively.

Randomisation

At the Baseline visit (Day -1), eligible subjects were randomized to treatment with 40 mg/day istradefylline or matching placebo in a 2:1 ratio. All subjects were randomized only once. The randomization scheme and codes is provided.

Blinding (masking)

The subject, site personnel, and the Sponsor were blinded to the treatment group to which a subject was assigned. Study monitors and data management personnel at KPI were unaware of the contents of the randomization code until after the study was completed, all data were validated, and the database was locked.

The istradefylline and placebo tablets used in this study were identical in appearance. Tablets were packaged in bottles identical in appearance and each containing 20 tablets.

The blind could be broken only if immediate knowledge of the study drug was needed to provide optimal clinical treatment to the subject. No unblinding occurred during the conduct of the study.

Statistical methods

The following analysis populations were used:

- The safety population was defined as all randomized subjects who received at least 1 dose of study drug.
- The ITT was defined as randomized subjects who received at least 1 dose of study drug, and who had both a baseline and at least 1 post-baseline measure for any of the efficacy variables.
- The modified ITT was defined as randomized subjects who received at least 1 dose of study drug, had any time spent in an OFF state at baseline, had both baseline and at least 1 post baseline measure for any of the efficacy variables, and were concordant in PD diary training.
- The efficacy-evaluable population (EFF) was defined as all randomized subjects who completed at least 2 weeks of treatment with no major protocol violations, had an average of at least 120 minutes of OFF time reported in 2 Baseline PD diaries, and at least 1 valid Endpoint diary. Major protocol violations included violation of inclusion/exclusion criteria, study drug non-compliance, protocol deviations, and inadequate PD diary training.

Analyses of the modified ITT and/or EFF populations, however, were to be carried out only if applicable.

The primary efficacy analysis was based on the ITT population, with a secondary analysis planned on the modified ITT population. Based on the results of the ITT population and possibly the modified ITT analyses, a supportive analysis was planned on the EFF population; however, data review, prior to

unblinding of the treatment groups, showed that data from 4 subjects who were randomized and received study drug were not evaluable for the modified ITT analysis. Since this would not have a relevant impact on outcome, the modified ITT analysis and the supportive analysis on the EFF population were not completed as proposed.

All efficacy analyses were carried out based on data from the ITT population.

For the purposes of the analysis method described below, a pooling strategy for Investigators was employed based on the enrollment summary of all subjects randomized for each Investigator. Investigators with less than 6 subjects randomized were pooled into 2 groups, based on geographic location: the South pool included 8 subjects randomized at Center 3 (5 subjects) and Center 17 (3 subjects); and the North pool included 9 subjects randomized at Center 25 (5 subjects), Center 11 (3 subjects), and Center 8 (1 subject). This pooling strategy was implemented prior to unblinding treatment codes.

The last observation carried forward (LOCF) approach was used to define the endpoint for efficacy variables. For each subject, this value was the week 12 value for each variable or the last available value at the time of study discontinuation. Actual values and change from baseline values were summarized for all efficacy variables at each time point collected and for the Endpoint.

All statistical tests were reported using an alpha level of 0.050. No adjustment for multiple testing was made. The Type I error rate was spent on the p-value for the primary efficacy variable of change from baseline in the percentage of awake time per day spent in an OFF state at Endpoint. If the primary efficacy variable showed statistical significance in favour of 40 mg/day istradefylline, the study was considered to have satisfied its objective.

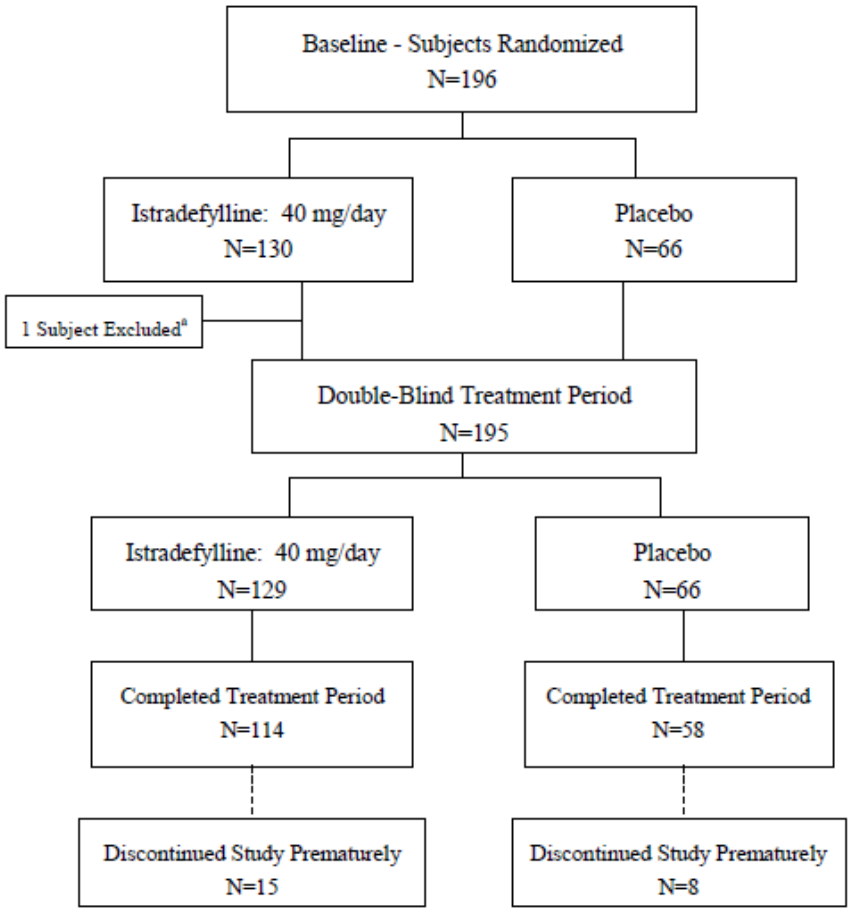
Interpretation of all p-values for the secondary efficacy variables was descriptive.

There were some changes to the analysis section mentioned in the study report prior to unblinding.

Results

Participant flow

Figure 25: Disposition of subjects



a One subject (Subject No. 22/162) was discontinued from the study prior to receiving the first does of study drug

Recruitment

Study locations: 22 centers in the US and 3 centers in Canada

Study period: first dose of study drug was on 23 April 2002 and last dose was 05 May 2003

Conduct of the study

Protocol amendments

The original protocol was issued on 30 January 2002. One amendment was made to the protocol (25 March 2002) prior to the first patient was dosed and introduced no major changes.

Protocol deviations

A total of 25 subjects did not meet all the protocol-specified entry criteria. According to the applicant, none of the deviations was considered to have a substantial effect on efficacy or safety analyses, and none resulted in the discontinuation of the subject's participation in the study.

Baseline data

Table 36: Demographic and other baseline characteristics (ITT population)

Demographic Characteristic	Placebo (N=66)	Istradefylline 40 mg/day (N=129)	Total (N=195)
Age (years)			
Mean	63.7	63.0	63.2
SD	10.06	8.98	9.34
Median	66.0	64.0	64.0
Range (min-max)	43 to 87	38 to 80	38 to 87
Sex (n [%])			
Male	40 (60.6)	77 (59.7)	117 (60.0)
Female	26 (39.4)	52 (40.3)	78 (40.0)
Race (n [%])			
Caucasian	61 (92.4)	124 (96.1)	185 (94.9)
Black	2 (3.0)	1 (0.8)	3 (1.5)
Asian	1 (1.5)	1 (0.8)	2 (1.0)
Hispanic	1 (1.5)	1 (0.8)	2 (1.0)
Other[a]	1 (1.5)	2 (1.6)	3 (1.5)
Height (cm)			
Mean	172.72	171.47	171.89
SD	9.213	9.885	9.657
Median	172.70	172.70	172.70
Range (min-max)	147.3 to 193.0	146.0 to 193.0	146.0 to 193.0
Weight[b] (kg)			
Mean	78.84	79.62	79.36
SD	20.212	18.751	19.209
Median	75.75	77.10	76.70
Range (min-max)	40.9 to 137.9	47.6 to 191.4	40.9 to 191.4

a "Other" included Portuguese, Pacific Islander, and Native American
b At screening
SD = standard deviation; min= minimum; max=maximum; cm=centimetre; kg=Kilogram

No notable differences were found between the istradefylline and placebo groups with respect to PD history. A summary of subjects' PD histories at Baseline is provided in Table 37. Both the mean time since PD diagnosis and the mean time since onset of motor complications were similar in both treatment groups.

Table 37: PD history at baseline (ITT population)

Characteristic	Placebo (N=66)	Istradefylline 40 mg/day (N=129)	Total (N=195)
Time since diagnosis (years)			
n[a]	44	75	119
Mean	9.27	9.30	9.29
SD	5.122	4.685	4.830
Median	7.95	8.50	8.40
Range (min to max)	1.4 to 19.1	2.4 to 25.6	1.4 to 25.6
Time since onset of motor complications (years)			
n[a]	44	77	121
Mean	3.57	3.29	3.39
SD	3.203	2.511	2.773
Median	2.35	2.70	2.50
Range (min to max)	0.2 to 15.2	0.1 to 11.9	0.1 to 15.2

Duration of PD history was calculated relative to the screening visit date
a Included only subjects who had no missing value for both the month and year of the event time
SD = standard deviation; min= minimum; max=maximum.

Both treatment groups were generally similar at Baseline for the primary efficacy variable of percentage of awake time per day spent in an OFF state. The 2 treatment groups were generally similar at Baseline

in the total hours of awake time per day spent in an OFF state and in the percentage and total hours of awake time per day spent in an ON state without dyskinesia and in an ON state without troublesome dyskinesia. The percentages and total hours of awake time per day in an ON state with dyskinesia (i.e., ON state with troublesome dyskinesia and ON state with non-troublesome dyskinesia combined), in an ON state with non-troublesome dyskinesia, and an ON state with troublesome dyskinesia were numerically lower in the istradefylline group in comparison with the placebo group.

Table 38: PD characteristics at baseline: percentage of awake time per day (ITT population)

Characteristic	Placebo (N=66)	Istradefylline 40 mg/day (N=129)	Total (N=195)
In an OFF state[a]			
Mean	37.19	38.44	38.02
SD	13.754	16.152	15.357
Median	35.35	36.90	36.20
Range (min to max)	14.4 to 70.0	0.0 to 92.8	0.0 to 92.8
In an ON state without dyskinesia[a]			
Mean	43.03	43.79	43.54
SD	23.482	20.799	21.687
Median	43.45	44.50	44.20
Range (min to max)	0.0 to 85.6	0.0 to 83.3	0.0 to 85.6
In an ON state with dyskinesia[a]			
Mean	19.78	17.77	18.45
SD	23.580	21.629	22.268
Median	6.65	7.10	7.10
Range (min to max)	0.0 to 82.8	0 to 100.0	0.0 to 100.0
In an ON state with non-troublesome dyskinesia[a]			
Mean	14.46	13.84	14.05
SD	18.346	18.005	18.076
Median	5.10	5.90	5.60
Range (min to max)	0.0 to 72.5	0.0 to 75.8	0.0 to 75.8
In an ON state with troublesome dyskinesia[a]			
Mean	5.32	3.93	4.40
SD	9.471	8.142	8.615
Median	0.00	0.00	0.00
Range (min to max)	0.0 to 35.3	0.0 to 43.1	0.0 to 43.1
In an ON state without troublesome dyskinesia[a]			
Mean	57.49	57.64	NA
SD	15.024	16.151	NA
Median	55.95	59.50	NA
Range (min to max)	27.7 to 85.6	0.0 to 88.6	NA

a Based on subjects' valid ON/OFF PD diaries from observed-case analysis
NA = not available; SD = Standard deviation; min= minimum; max=maximum.

Table 39: PD characteristics at baseline: total hours of awake time per day (ITT population)

Characteristic	Total Hours of Awake Time per Day Placebo (N=66)	Istradefylline 40 mg/day (N=129)
In an OFF state[a]		
Mean	6.20	6.37
SD	2.457	2.724
Median	5.80	6.0
Range (min to max)	2.0 to 12.3	0.0 to 14.5
In an ON state without dyskinesia[a]		
Mean	7.06	7.25
SD	3.815	3.519
Median	7.05	7.30
Range (min to max)	0.0 to 15.0	0.0 to 15.0
In an ON state with dyskinesia[a]		
Mean	3.34	2.92
SD	3.966	3.585
Median	1.15	1.30
Range (min to max)	0 to 13.3	0.0 to 16.3
In an ON state with non-troublesome dyskinesia[a]		
Mean	2.43	2.29
SD	3.054	3.022
Median	0.90	0.80
Range (min to max)	0.0 to 13.0	0.0 to 13.5
In an ON state with troublesome dyskinesia[a]		
Mean	0.91	0.64
SD	1.617	1.305
Median	0.00	0.00
Range (min to max)	0.0 to 6.3	0.0 to 7.0
In an ON state without troublesome dyskinesia[a]		
Mean	9.50	9.54
SD	2.599	2.809
Median	9.50	10.00
Range (min to max)	3.0 to 15.3	0.0 to 15.6

a Based on subjects' valid ON/OFF PD diaries from observed-case analysis
NA = not available; SD = Standard deviation; min= minimum; max=maximum.

In addition to the use of levodopa and carbidopa, all but 1 subject received other prior medications within 30 days prior to the first dose of DB study drug that were either stopped prior to randomization or continued during the double-blind treatment period. As expected in this population, the majority (85.6%) of subjects received prior treatment with dopaminergic agents. For subjects in the safety population, the next most frequently used prior therapeutic classes were the analgesics and antipyretic agents, anti-inflammatory/anti-rheumatic products, non-steroids, or antidepressants. Most subjects in both groups continued taking these medications during the DB treatment period.

The most frequently reported concomitant medications, excluding levodopa and carbidopa, by therapeutic class in both treatment groups were dopaminergic agents, other analgesics and antipyretic agents, anti-inflammatory/antirheumatic products, non-steroids, and antidepressants. In general, most subjects in both groups received concomitant medications agents as prior therapy and continued taking these medications during the DB treatment period, often receiving the same medications with no additional concomitant medications added after randomization. 100% of patients in both groups were taking prior L/C therapy

Numbers analysed

Table 40: Subject disposition and study populations

Status	Placebo (N=66) n (%)	Istradefylline 40 mg/day (N=130) n (%)
All Subjects Randomized	66 (100.0)	130 (100.0)[a]
Safety Population[b]	66 (100.0)	129 (99.2)
Intent-to-Treat Population[c]	66 (100.0)	129 (99.2)
Completed		
Week 2	64 (97.0)	126 (96.9)
Week 4	59 (89.4)	120 (92.3)
Week 8	58 (87.9)	117 (90.0)
Completed Double-blind Treatment Period	58 (87.9)	114 (87.7)
Discontinued Study Prematurely	8 (12.1)	16 (12.3)
Reason for Discontinuation from Study		
Lack of efficacy	0 (0.0)	1 (0.8)
Adverse events	5 (7.6)	10 (7.7)
Protocol violation or non-compliance with study drug	0 (0.0)	1 (0.8)
Subject withdrew consent	3 (4.5)	4 (3.1)

a One subject experienced AEs prior to receiving study drug and was discontinued from the study. This subject was not included in any efficacy or safety analyses.
b All randomised subjects who received at least 1 dose of study drug
c All randomised subjects who received at least 1 dose of study drug and had a baseline and at least 1 post-baseline efficacy measurement.

Outcomes and estimation

Primary efficacy variable

The primary efficacy variable was the change from baseline in the percentage of awake time per day spent in an OFF state at endpoint based on the subjects' valid ON/OFF PD diary data (observed case analysis).

The mean percentages of awake time per day spent in an OFF state at baseline were 38.44% for the 40 mg/day istradefylline group and 37.19% for the placebo group. At Endpoint, these values were 27.40% and 32.65%, respectively, for the 2 groups. Thus, for subjects in the istradefylline group, a greater mean decrease (-10.81%) from baseline to endpoint was observed in the percentage of awake time per day spent in an OFF state compared to that in the placebo group (-4.04%) and this difference was statistically significant (p-value = 0.007) based on the LSM from the ANOVA model. The analysis of covariance (ANCOVA) analysis showed a similar result (p-value = 0.007).

Table 41: Change from baseline at endpoint in percentage of awake time per day spent in an OFF state (observed case analysis)

	Percentage of Awake Time Per Day Spent in an OFF State[a]		
Visit	Placebo (N=66)	Istradefylline 40 mg/day (N=129)	p-value[b]
Baseline			
n	66	129	
Mean	37.19	38.44	
SD	13.754	16.152	
Median	35.35	36.90	
Min to max	14.4 to 70.0	0 to 92.8	
Endpoint			
n	65	126	
Mean	32.65	27.40	
SD	16.192	16.887	
Median	33.90	25.85	
Min to max	0.0 to 60.2	0 to 79.8	
Change from Baseline at Endpoint			
n	65	126	
Mean	-4.04	-10.81	0.007
SD	15.743	16.556	
Median	-4.20	-10.55	
Min to max	-49.1 to 25.6	-50.4 to 47.5	

a Based on subjects valid ON/OFF PD diaries from observed-case analysis
b The p-value based on LSM from 2-way ANOVA, including terms for investigator and treatment
NA = not available; SD = Standard deviation; min= minimum; max=maximum.

Table 42: Sensitivity Analyses of the Primary Endpoint (reduction in daily off time)

Analysis Method		Statistic	Placebo	Istradefylline 40 mg/day
WORST CASE				
		Baseline[a] (mean)	37.47%	38.54%
		Endpoint[a] (mean)	32.86%	27.73%
ANOVA	WC	LS mean change from baseline to endpoint	-3.80%	-10.27%
		LS mean difference from placebo (95% CI)	-	-6.47% (-11.30, -1.64)
ANCOVA	WC	LS mean change from baseline to endpoint	-4.38%	-10.32%
		LS mean difference from placebo (95% CI)	-	-5.94% (-10.34, -1.54)
HOURS PER DAY				
ANOVA	OC	Baseline[a] (mean)	6.20 h	6.37 h
		Endpoint[a] (mean)	5.49 h	4.55 h
		LS mean change from baseline to endpoint	-0.60 h	-1.76 h
		LS mean difference from placebo (95% CI)	-	-1.16 h (-1.98, -0.34)
	WC	Baseline[a] (mean)	6.27 h	6.42 h
		Endpoint[a] (mean)	5.56 h	4.63 h
		LS mean change from baseline to endpoint	-0.61 h	-1.73 h
		LS mean difference from placebo (95% CI)	-	-1.12 h (-1.93, -0.30)
ANCOVA	OC	LS mean change from baseline to endpoint	-0.67 h	-1.75 h
		LS mean difference from placebo (95% CI)	-	-1.08 h (-1.82, -0.34)
	WC	LS mean change from baseline to endpoint	-0.68 h	-1.73 h
		LS mean difference from placebo (95% CI)	-	-1.05 h (1.79, -0.31)

Analysis Method		Statistic	Placebo	Istradefylline 40 mg/day
SCE ANALYSIS				
MMRM	OC	Baseline[a] (mean)	6.20 h	6.37 h
		Week 12 (mean)	5.45 h	4.41 h
		LS mean change from baseline to Week 12	-0.67 h	-1.78 h
		LS mean difference from placebo (95% CI)	-	-1.11 h (-1.87, -0.35)
	WC	Baseline[a] (mean)	6.20 h	6.42 h
		Week 12 (mean)	5.45 h	4.49 h
		LS mean change from baseline to Week 12	-0.69 h	-1.75 h
		LS mean difference from placebo (95% CI)	-	-1.06 h (-1.82, -0.30)

a Baseline and endpoint values for OC and WC (for both percentage and hours) were the same irrespective of the analysis that was performed

NA=not applicable; ANCOVA=Analysis of covariance; ANOVA=Analysis of variance; CI=Confidence interval; LS= Least squares; MMRM=Mixed-model repeated measures; OC=Observed case; SCE=Summary of clinical efficacy; WC=Worst Case.

Secondary Efficacy variables

Several analysis were provided by the applicant for secondary efficacy variables. Overall, secondary efficacy endpoints were in favour of istradefylline. In particular, change from baseline to endpoint in total hours spent in OFF time per day for istradefylline compared to placebo was performed as a secondary endpoint. The difference between treatment groups of 1.15 hours less time spent in an OFF state for the istradefylline 40mg dose compared to placebo could be considered clinically relevant for patients. Additionally, the percentage of awake time/day spent ON without troublesome dyskinesia as another clinically relevant secondary endpoint showed changes favouring istradefylline (see below table).

Due to the lack of multiplicity control, results for these secondary efficacy variables were solely descriptive.

Summary of the study

This summary should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 43: Summary of efficacy for trial 6002-US-005

Title: A 12-Week, Double-Blind, Placebo-Controlled, Randomized, Multicenter Study of the Efficacy of 40 mg/day KW-6002 as Treatment for Parkinson's Disease in Patients with Motor Response Complications on Levodopa/Carbidopa Therapy			
Study identifier	6002-US-005		
Design	Phase 2b, randomised, double-blind, placebo-controlled, fixed-dose, parallel-group, multicenter trial		
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:		12 weeks Not applicable Not applicable
Hypothesis	Superiority		
Treatments groups	Istradefylline 40 mg		Istradefylline 40 mg once daily, 12 weeks, n=130 subjects
	Placebo		Placebo once daily, 12 weeks, n=66 subjects
Endpoints and definitions	Primary endpoint	OFF time per day	Change from baseline in the percentage of awake time/day spent in the OFF state
	Key secondary endpoint	ON time per day without troublesome dyskinesia	Change from baseline in percentage of awake time/day spent ON without troublesome dyskinesia
Database lock	23 June 2003		
Results and Analysis			
Analysis description	**Primary Analysis**		
Analysis population and time point description	Intent to treat (ITT), week 12/Endpoint LOCF, Observed case, ANOVA		

Descriptive statistics and estimate variability	Treatment group	Placebo	Istradefylline 40 mg
	Number of subjects analysed at Week 12/Endpoint	65	126
	Change from baseline in the percentage of awake time/day spent in the OFF state; Mean (SD)	-4.04 (15.743)	-10.81 (16.556)
	Range (min to max)	-49.1 to 25.6	-50.4 to 47.5
	Change from baseline in percentage of awake time/day spent in the ON state without troublesome dyskinesia; Mean (SD)	3.62 (17.439)	9.22 (17.527)
	Range (min to max)	-42.5 to 49.1	-44.3 to 48.7
Effect estimate per comparison	<u>Primary endpoint</u> (% of awake time/day spent in the OFF state); difference from placebo in the LSM change from baseline	Comparison groups	Istradefylline vs Placebo
		LSM change vs. placebo	-6.78
		95% CI	-11.63, -1.92
		P-value	0.007
	<u>Key secondary endpoint</u> (% of awake time/day spent ON without troublesome dyskinesia); difference from placebo in the LSM change from baseline	Comparison groups	Istradefylline vs Placebo
		LSM change vs. placebo	5.60
		95% CI	0.37, 10.83
Analysis description	**Sensitivity analysis,** ITT, week 12/Endpoint, LOCF, Observed case, ANCOVA		
	% of awake time spent in the OFF state, LSM difference from placebo in the change from baseline [95%CI]: Istradefylline 40 mg, -6.17 [-10.60, -1.74]		

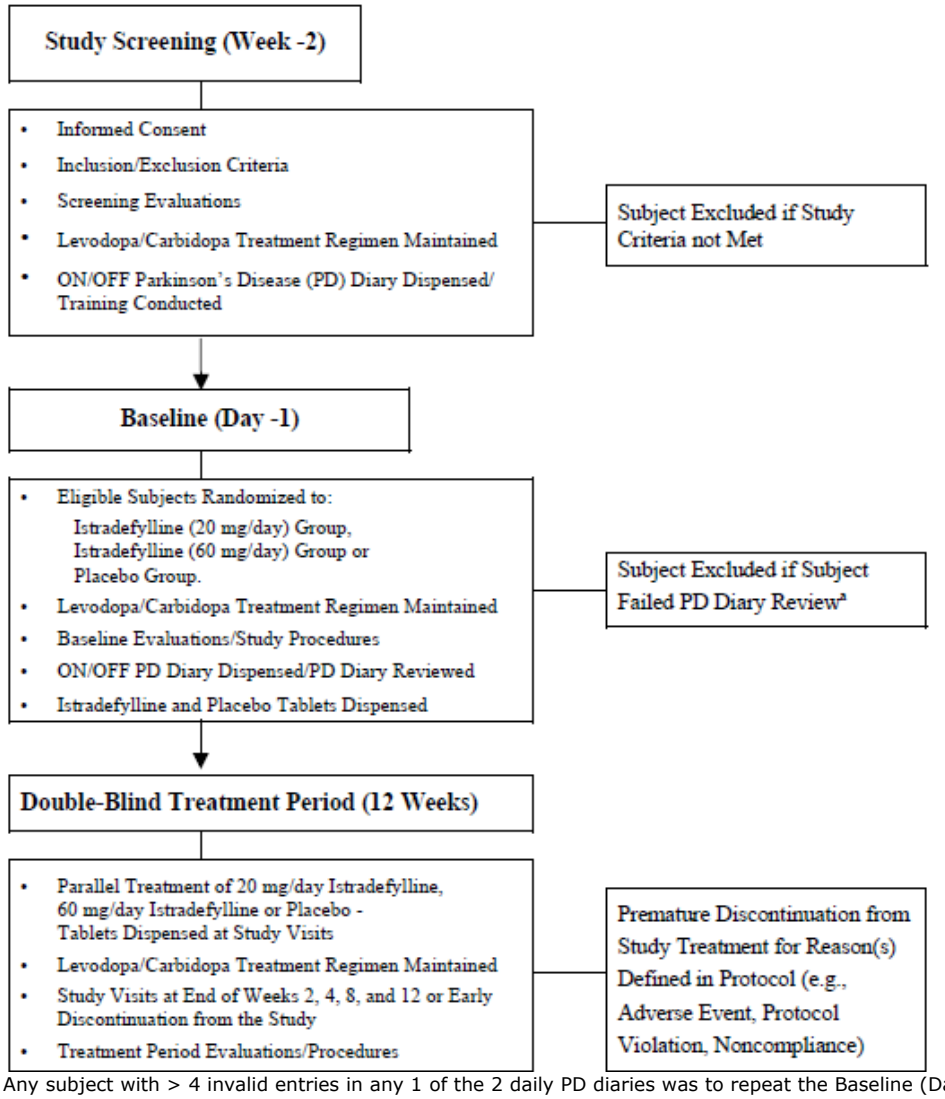
Study 6002-US-006

"A 12-Week, Double-Blind, Placebo-Controlled, Randomized, Multicenter Study of the Efficacy of Doses of 20 and 60 mg/day KW-6002 as Treatment for Parkinson's Disease in Patients With Motor Response Complications on Levodopa/Carbidopa Therapy"

Methods

Study Design

Figure 26: Study plan of Study 6002-US-006



Any subject with > 4 invalid entries in any 1 of the 2 daily PD diaries was to repeat the Baseline (Day-1) visit 7 days later. A subject who failed the study entry criteria for diary validation a second time was excluded from participating in the study.

Study Participants

Inclusion and exclusion criteria were the same as for study 6002-US-005.

Treatments

The study treatments were 20 mg of istradefylline, 60 mg of istradefylline or matching placebo. Subjects took 2 tablets of study drug each day as follows:

- Subjects in the placebo group took one 20-mg matching placebo tablet plus one 40-mg matching placebo tablet.
- Subjects in the 20 mg/day istradefylline group took one 20-mg istradefylline tablet plus one 40-mg matching placebo tablet.
- Subjects in the 60 mg/day istradefylline group took one 20-mg istradefylline tablet plus one 40-mg istradefylline tablet.

During the study, subjects were to take L/C according to the Investigator's clinical judgment and the requirements of the protocol. In addition, other antiparkinson medications were permitted.

Objectives

Primary Objective

The primary objective of this study was to evaluate the safety and establish the efficacy of 20mg/day and 60 mg/day istradefylline for reducing OFF time in subjects with advanced PD treated with L/C.

Secondary Objective

The secondary objective of this study was to establish the efficacy of 20 and 60 mg/day istradefylline for reducing motor symptoms and improving ADL in subjects with advanced PD treated with L/C.

Outcomes/endpoints

Primary Efficacy Measurements and Endpoints

The primary efficacy variable of this study was the mean change from Baseline in the percentage of awake time per day spent in the OFF state at Endpoint (Week 12 value or the last available post-baseline value at the time of premature discontinuation from the study) based on the subjects' valid ON/OFF PD diary.

To meet this objective, subjects recorded assessments of ON and OFF time every 30 minutes during awake time for 24 hours in the subjects' ON/OFF PD diary.

Secondary Efficacy Measurements and Endpoints

Secondary efficacy measurements were based on the changes in the subjects' valid ON/OFF PD diary data, the UPDRS scale, and the CGI-I scale. The definition of ON and OFF times for the secondary efficacy variables was the same as that provided for the primary efficacy variable. (See 6002-US-005)

Sample size

The sample sizes of the treatment groups were based on the expected difference between treatment groups in the percentage of time OFF from Baseline to Endpoint for the ITT population. A total of 130 subjects in each of the istradefylline groups were planned to be randomized to obtain at least 112 subjects in each istradefylline group. For the placebo group, 65 subjects were planned to be randomized to obtain at least 56 subjects in the ITT population. The sample size of each group was sufficient to provide 80% power to detect statistical significance at the 2-sided alpha level of 0.050 for treatment group differences greater than or equal to 50% of the applicable SD.

Randomization

At the Baseline visit (Day -1), eligible subjects were randomized to treatment with 20 mg/day istradefylline, 60 mg/day istradefylline, or matching placebo in a 2:2:1 ratio according to a randomization schedule. All subjects were randomized only once. The randomization scheme and codes is provided.

Blinding (masking)

During the clinical conduct of the study (i.e., prior to the last subject dosing on 06 October 2003), study drug was unblinded for one subject in the 20 mg/day istradefylline group at the request of the Investigator because of a Serious AE (SAE) (cardio-respiratory arrest and loss of consciousness).

Following an administrative letter sent to all Investigators describing new non-clinical safety findings (07 October 2003), study drug was unblinded for another subject in the 20 mg/day istradefylline group at the request of the Investigator. The unblinding occurred after the final statistical analysis plan (SAP) was signed on 2 October 2003 and prior to the unblinding of the study database (30 November 2003).

Statistical methods

The SAP for this study was developed and finalized prior to the unblinding of treatment codes. The final SAP was signed on 2 October 2003, and the database was unblinded for the efficacy analyses on 30 November 2003.

The following subject populations were used safety population, ITT, modified ITT and EFF as defined for study 6002-US-005.

Analyses of the modified ITT and/or EFF populations, however, were to be carried out only if applicable.

All efficacy analyses were carried out based on data from the ITT population. In addition, analyses of the change from Baseline by study visit and at Endpoint in percentage and total hours of awake time per day spent in an OFF state were carried out based on the modified ITT population using both the observed-case and worst-case imputation approaches. The supportive analysis on the EFF population was not conducted as proposed.

For purposes of the analysis method describe below, a pooling strategy for Investigators was employed based on the enrollment summary of all subjects randomized for each Investigator. Investigators with less than 10 subjects randomized were pooled into 6 groups based on geographic location (Northeast, Midwest 1, Midwest 2, Midwest 3, Southwest, and Southeast). This pooling strategy was implemented prior to unblinding treatment codes.

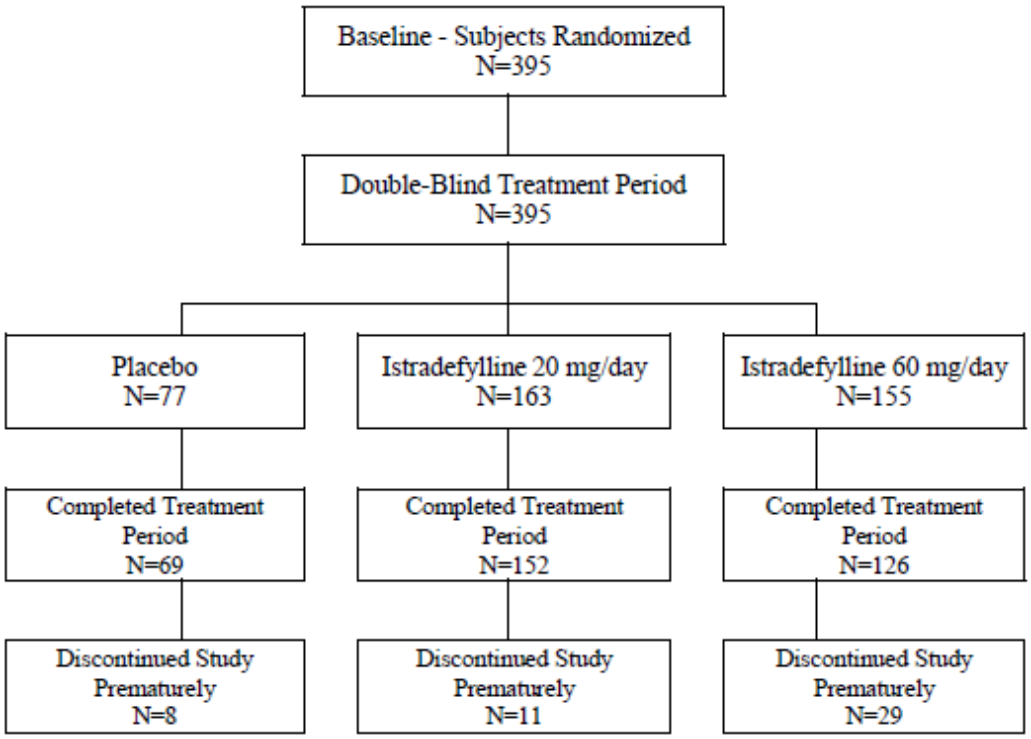
The LOCF approach was used to define the endpoint for efficacy variables. For each subject, this value was the Week 12 value for each variable or the last available post-baseline value at the time of study discontinuation. Actual values and change from Baseline values were analysed for all efficacy variables (except CGI-I) at each time point collected and for the Endpoint.

All efficacy variables (except CGI-I) were analysed using the ANOVA. An ANCOVA was used as a supportive analysis for testing the treatment effects as specified in the SAP. The ANOVA model contained terms for Investigator and treatment. The ANCOVA model contained terms for Investigator, treatment, and Baseline as a covariate. In the context of the ANOVA and ANCOVA models described above, a closed testing procedure was used to test for treatment effects. First, the overall test for treatment effects versus placebo was carried out using the F-test with 2 degrees of freedom for the numerator from the Type III sums of squares in the ANOVA or ANCOVA model described above using a significance level of 0.050. If this test was significant, the pairwise comparisons of each of the least squares means of the 2 dose groups with placebo was carried out using a significance level of 0.050. If both tests were significant, both dose groups were declared simultaneously significantly different from placebo. If only 1 of the 20 or 60 mg/day istradefylline dose groups was significantly different from placebo, the respective group was declared individually different from placebo. This testing procedure controlled the experiment-wise significance level at 0.050. Interpretation of all p-values for secondary efficacy variables was descriptive.

Results

Participant flow

Figure 27: Study Participant flow



Recruitment

Study locations: 38 centers in the US and 2 centers in Canada

Study period: first dose of study drug was on 23 April 2002 and last dose was 06 October 2003

Conduct of the study

Amendments

The original protocol was issued on 30 January 2002. Three amendments were made to the protocol: Amendment 1 (05 April 2002); Amendment 2 (18 September 2002); and Amendment 3 (26 November 2002).

The first amendment was made prior to study initiation. Amendment 2 involved change of CRO, to manage increasing administrative needs of programme. Amendment 3 included further clarification of subject diary training and documentation of this process.

Protocol Deviations

A total of 108 subjects did not meet all protocol-specified entry criteria. According to the applicant, none of the deviations was considered to have a substantial effect on efficacy or safety analyses, and none resulted in the discontinuation of the subject's participation in the study or the exclusion of the subject's data from the analyses based on the ITT or Safety populations.

Baseline data

Demographic characteristics of age, race, sex, height, and weight were similar among the 3 groups.

Table 44: Demographic and other baseline characteristics (ITT population)

Demographic Characteristic	Placebo (N=77)	Istradefylline 20 mg/day (N=163)	Istradefylline 60 mg/day (N=155)	Total (N=395)
Age (yrs)				
Mean	63.0	65.0	63.5	64.0
SD	12.05	9.59	10.08	10.31
Median	63.0	66.0	65.0	65.0
Range (min-max)	38 to 87	36 to 87	36 to 83	36 to 87
Sex (n [%])				
Male	54 (70.1)	104 (63.8)	106 (68.4)	264 (66.8)
Female	23 (29.9)	59 (36.2)	49 (31.6)	131 (33.2)
Race (n [%])				
Caucasian	73 (94.8)	151 (92.6)	141 (91.0)	365 (92.4)
Black	0	2 (1.2)	4 (2.6)	6 (1.5)
Asian	1 (1.3)	7 (4.3)	2 (1.3)	10 (2.5)
Hispanic	1 (1.3)	1 (0.6)	4 (2.6)	6 (1.5)
American Indian	0	1 (0.6)	0	1 (0.3)
Other[a]	2 (2.6)	1 (0.6)	4 (2.6)	7 (1.8)
Height (cm)			(n=154) [b]	(n=394)[b]
Mean	170.81	170.70	172.84	171.56
SD	10.371	11.166	10.476	10.769
Median	172.70	171.50	174.50	172.70
Range (min-max)	145.0 to 198.0	140.2 to 200.7	152.4 to 203.2	140.2 to 203.2
Weight[c] (kg)				
Mean	80.83	75.08	78.86	77.68
SD	16.316	16.981	16.782	16.890
Median	82.10	73.90	79.40	77.60
Range (min-max)	49.7 to 127.5	42.4 to 130.6	43.5 to 133.0	42.4 to 133.0

a "Other" included Guyanese, Indian, Middle Eastern, Portuguese, Asian Indian, Haitian and English.
B For the 60mg/day istradefylline group, the site reported height for subject No. 79/132 as "not done"
c At screening
SD = standard deviation; min= minimum; max=maximum; cm=centimetre; kg=Kilogram

No notable differences were found among the 3 groups with respect to PD history.

Table 45: PD history at baseline (ITT population)

Characteristic	Placebo (N=77)	Istradefylline 20 mg/day (N=163)	Istradefylline 60 mg/day (N=155)	Total (N=395)
Time since diagnosis (years)				
n[a]	54	98	95	247
Mean	8.69	9.24	7.94	8.62
SD	5.002	5.275	4.017	4.781
Median	7.80	8.75	7.40	8.20
Range (min to max)	2.4 to 32.3	2.1 to 31.9	1.7 to 16.2	1.7 to 32.3
Time since onset of motor complications (years)				
n[a]	40	74	84	198
Mean	3.81	3.71	3.27	3.54
SD	2.694	3.830	3.035	3.288
Median	2.95	2.55	2.30	2.55
Range (min-max	0.4 to 9.3	0.1 to 26.2	0.2 to 15.8	0.1 to 26.2

Duration of PD history was calculated relative to the screening visit date
a Included only subjects who had no missing value for both the month and year of the event time
SD = standard deviation; min= minimum; max=maximum.

Table 46: PD characteristics at baseline Percentage of awake time per day (ITT population)

	Percentage of Awake Time per Day			
Characteristic	Placebo (N=77)	Istradefylline 20 mg/day (N=163)	Istradefylline 60 mg/day (N=155)	Total (N=395)
In an OFF state[a]				
Mean	36.56	34.81	35.07	35.25
SD	14.336	14.701	13.748	14.241
Median	36.90	32.40	35.10	34.20
Range (min to max)	4.2 to 81.4	8.2 to 82.5	6.2 to 72.4	4.2 to 82.5
In an ON state without dyskinesia[a]				
Mean	44.15	42.60	47.83	44.96
SD	19.524	22.388	18.82	20.586
Median	47.20	43.60	50.00	46.30
Range (min-max)	0.0 to 82.8	0.0 to 86.4	0.0 to 87.5	0.0 to 87.5
In an ON state with dyskinesia[a]				
Mean	19.29	22.59	17.10	19.79
SD	20.325	22.001	19.789	20.927
Median	14.10	18.90	9.4	13.60
Range (min-max)	0.0 to 94.4	0.0 to 81.8	0.0 to 73.2	0.0 to 94.4
In an ON state with non-troublesome dyskinesia[a]				
Mean	16.13	16.82	13.57	15.41
SD	18.567	17.233	15.512	16.879
Median	10.60	13.30	9.00	11.20
Range (min-max)	0.0 to 94.4	0.0 to 74.2	0.0 to 64.0	0.0 to 94.4
In an ON state with troublesome dyskinesia[a]				
Mean	3.17	5.77	3.53	4.38
SD	5.556	11.435	7.728	9.189
Median	0.00	0.00	0.00	0.00
Range (min-max)	0.0 to 22.6	0.0 to 62.5	0.0 to 37.8	0.0 to 62.5
In an ON state without troublesome dyskinesia[a]				
Mean	60.06	59.11	61.22	NA
SD	15.384	16.688	13.940	NA
Median	58.80	61.30	62.30	NA
Range (min-max)	18.6 to 95.8	10.3 to 87.1	27.6 to 92.3	NA

a Based on subjects valid ON/OFF PD diaries from observed-case analysis
NA = not available; SD = Standard deviation; min= minimum; max=maximum.

Table 47: PD characteristics at baseline total hours of awake time per day (ITT population)

	Hours of Awake Time Per Day		
Characteristics	Placebo (N=77)	Istradefylline 20 mg/day (N=163)	Istradefylline 60 mg/day (N=155)
In an OFF state[a]			
Mean	6.31	5.72	5.81
SD	2.591	2.502	2.339
Median	6.30	5.30	5.80
Range (min to max)	0.8 to 12.3	1.3 to 14.8	1.0 to 13.0
In an ON state without dyskinesia[a]			
Mean	7.55	6.97	7.89
SD	3.449	3.723	3.201
Median	7.80	7.00	7.80
Range (min-max)	0.0 to 14.5	0.0 to 14.0	0.0 to 15.5
In an ON state with dyskinesia[a]			
Mean	3.28	3.73	2.85
SD	3.437	3.720	3.299
Median	2.30	3.30	1.50
Range (min-max)	0.0 to 16.5	0.0 to 14.5	0.0 to 12.3
In an ON state with non-troublesome dyskinesia[a]			
Mean	2.74	2.75	2.26
SD	3.140	2.799	2.587
Median	1.80	2.00	1.50
Range (min-max)	0.0 to 6.5	0.0 to 12.3	0.0 to 10.8
In an ON state with troublesome dyskinesia[a]			
Mean	0.55	1.00	0.59
SD	0.935	2.066	1.302
Median	0.00	0.00	0.00
Range (min-max)	0.0 to 4.0	0.0 to 11.8	0.0 to 6.3
In an ON state without troublesome dyskinesia[a]			
Mean	10.29	9.72	10.16
SD	2.698	2.836	2.475
Median	10.50	10.00	10.30
Range (min-max)	2.8 to 16.8	1.8 to 15.3	4.3 to 16.0

a Based on subjects valid ON/OFF PD diaries from observed-case analysis
SD = Standard deviation; min= minimum; max=maximum.

In addition to the use of levodopa and carbidopa, all but 1 subject received other prior and concomitant medications within 30 days prior to the first dose of DB study drug that either stopped prior to randomization or continued during the DB treatment period. As expected in this population, the majority (90.9%) of subjects received prior treatment with dopaminergic agents. The most commonly reported concomitant medications, excluding levodopa and carbidopa, by therapeutic class among all subjects were dopaminergic agents, other analgesics and antipyretic agents, non-steroidal antiinflammatory/ antirheumatic products, and other plain vitamin preparations. In general, most subjects in all 3 groups received concomitant medications agents as prior therapy and continued taking these medications during the DB treatment period, often receiving the same medications with no additional concomitant medications added after randomization.

Numbers analysed

Table 48: Subject disposition

Status	Placebo (N=77) n (%)	Istradefylline 20 mg/day (N=163) n (%)	Istradefylline 60 mg/day (N=155) n (%)
All Subjects Randomized:	77 (100.0)	163 (100.0)	155 (100.0)
Safety Population[a]	77 (100.0)	163 (100.0)	155 (100.0)
Intent-to-Treat Population[b]	77 (100.0)	163 (100.0)	155 (100.0)
Completed:			
Week 2	74 (96.1)	162 (99.4)	144 (92.9)
Week 4	72 (93.5)	157 (96.3)	137 (88.4)
Week 8	70 (90.9)	153 (93.9)	132 (85.2)
Completed Double-blind Treatment Period	69 (89.6)	152 (93.3)	126 (81.3)
Discontinued Study Prematurely	8 (10.4)	11 (6.7)	29 (18.7)
Reason for Discontinuation from Study			
Lack of efficacy	0	1 (0.6)	2 (1.3)
Adverse event	5 (6.5)	6 (3.7)	16 (10.3)
Protocol violation or non-compliance with study drug	2 (2.6)	2 (1.2)	5 (3.2)
Subject withdrew consent	1 (1.3)	2 (1.2)	3 (1.9)
Other	0	0	3 (1.9)

a All randomised subjects who received at least 1 dose of study drug
b All randomised subjects who received at least 1 dose of study drug and had a baseline and at least 1 post-baseline efficacy measurement.

Outcomes and estimation

Primary Efficacy Variable

Table 49: Percentage of awake time per day spent in an OFF state (Observed Case Analysis)

	Percentage of Awake Time Per Day Spent in an OFF State[a]			
Visit	Placebo (N=77)	Istradefylline 20 mg/day (N=163)	Istradefylline 60 mg/day (N=155)	Overall p-value[b]
Baseline				
n	77	163	155	
Mean	36.56	34.81	35.07	
SD	14.336	14.701	13.748	
Median	36.90	32.40	35.10	
Min to max	4.2 to 81.4	8.2 to 82.5	6.2 to 72.4	
Endpoint				
n	75	162	148	
Mean	32.09	26.87	26.80	
SD	18.328	15.376	15.263	
Median	33.50	25.70	25.80	
Min to max	0.0 to 89.1	0.0 to 76.0	0.0 to 75.7	
Change from Baseline at Endpoint				
n	75	162	148	
Mean	-4.27	-7.94	-8.13	
SD	17.214	15.518	14.164	
Median	-3.90	-7.35	-8.65	
Min to max	-50.1 to 50.6	-67.9 to 32.7	-56.8 to 51.9	
ANOVA				0.169
LS mean	-4.07	-7.72	-7.84	
LS mean difference (versus placebo)		-3.65	-3.77	
p-value (versus placebo)[c]		0.088	0.082	
ANCOVA				0.049
LS mean	-3.47	-7.83	-7.96	
LS mean difference (versus placebo)		-4.36	-4.49	
p-value (versus placebo)[c]		0.026	0.024	

a Based on subjects' valid ON/OFF PD diaries from observed-case analysis
b The overall p-value based on Type III Sums of squares F-test with 2 degrees of freedom for ANOVA or ANCOVA
c P-value for individual comparisons (istradefylline groups versus placebo group) based on LSM from 2-way ANOVA including terms for investigator and treatment or ANCOVA, including terms for baseline, investigator and treatment. At endpoint, a closed testing procedure was used for treatment effects
LSM = Least squares mean; SD = Standard deviation; min= minimum; max=maximum.

Secondary Efficacy variables

In failing to meet the primary endpoint, by prespecified analysis method, Study 6002-US-006 is considered a formally failed study. Therefore, results for secondary efficacy endpoints are not further considered (percentage of awake time / day spent ON without troublesome dyskinesia is provided in the table below for completeness as a relevant secondary efficacy endpoint).

Summary of main efficacy results

This summary should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 50: Summary of efficacy for trial 6002-US-006

Title: A 12-Week, Double-Blind, Placebo-Controlled, Randomized, Multicenter Study of the Efficacy of Doses of 20 and 60 mg/day KW-6002 as Treatment for Parkinson's Disease in Patients With Motor Response Complications on Levodopa/Carbidopa Therapy					
Study identifier	6002-US-006				
Design	Phase 2b, randomised, double-blind, placebo-controlled, fixed-dose, parallel-group, multicenter trial				
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:		12 weeks Not applicable Not applicable		
Hypothesis	Superiority				
Treatments groups	Istradefylline 20 mg		Istradefylline 20 mg once daily, 12 weeks, n=163 subjects		
	Istradefylline 60 mg		Istradefylline 60 mg once daily, 12 weeks, n=155 subjects		
	Placebo		Placebo once daily, 12 weeks, n=77 subjects		
Endpoints and definitions	Primary endpoint	OFF time per day	Change from baseline in the percentage of awake time/day spent in the OFF state		
	Key secondary endpoint	ON time per day without troublesome dyskinesia	Change from baseline in percentage of awake time/day spent ON without troublesome dyskinesia		
Database lock	20 November 2003				
Results and Analysis					
Analysis description	**Primary Analysis**				
Analysis population and time point description	Intent to treat (ITT), week 12/Endpoint LOCF, Observed case, ANOVA				
Descriptive statistics and estimate variability	Treatment group		Placebo	Istradefylline 20 mg	Istradefylline 60 mg
	Number of subjects analysed at Week 12/ Endpoint		75	162	148
	Change from baseline in the percentage of awake time/day spent in the OFF state; Mean (SD)		-4.27 (17.214)	-7.94 (15.518)	-8.13 (14.164)
	Range (min to max)		-50.1 to 50.6	-67.9 to 32.7	-56.8 to 51.9
	Change from baseline in percentage of awake time/day spent in the ON state without troublesome dyskinesia; Mean (SD)		3.54 (18.141)	7.28 (16.771)	7.19 (15.577)
	Range (min to max)		-50.6 to 50.1	-50.5 to 58.5	-51.9 to 56.8
Effect estimate per comparison	Primary endpoint (% of awake time/day spent in the OFF state); difference from placebo		Comparison groups	Istradefylline 20mg vs Placebo	Istradefylline 60mg vs Placebo

	in the LSM change from baseline	LSM change vs. placebo	-3.65	-3.77
		95% CI	-7.83, 0.53	-8.01, 0.47
		P-value	0.088	0.082
	<u>Key secondary endpoint</u> (% of awake time/day spent ON without troublesome dyskinesia); difference from placebo in the LSM change from baseline	Comparison groups	Istradefylline 20mg vs Placebo	Istradefylline 60mg vs Placebo
		LSM change vs. placebo	3.89	3.55
		95% CI	-0.68, 8.46	-1.08, 8.19
Analysis description	**Sensitivity analysis,** ITT, week 12/Endpoint, LOCF, Observed case, ANCOVA			
	% of awake time spent in the OFF state, LSM difference from placebo in the change from baseline [95%CI]: Istradefylline 20mg, -4.35 [-8.16, -0.54]; Istradefylline 60mg, -4.49 [-8.35, -0.62]			

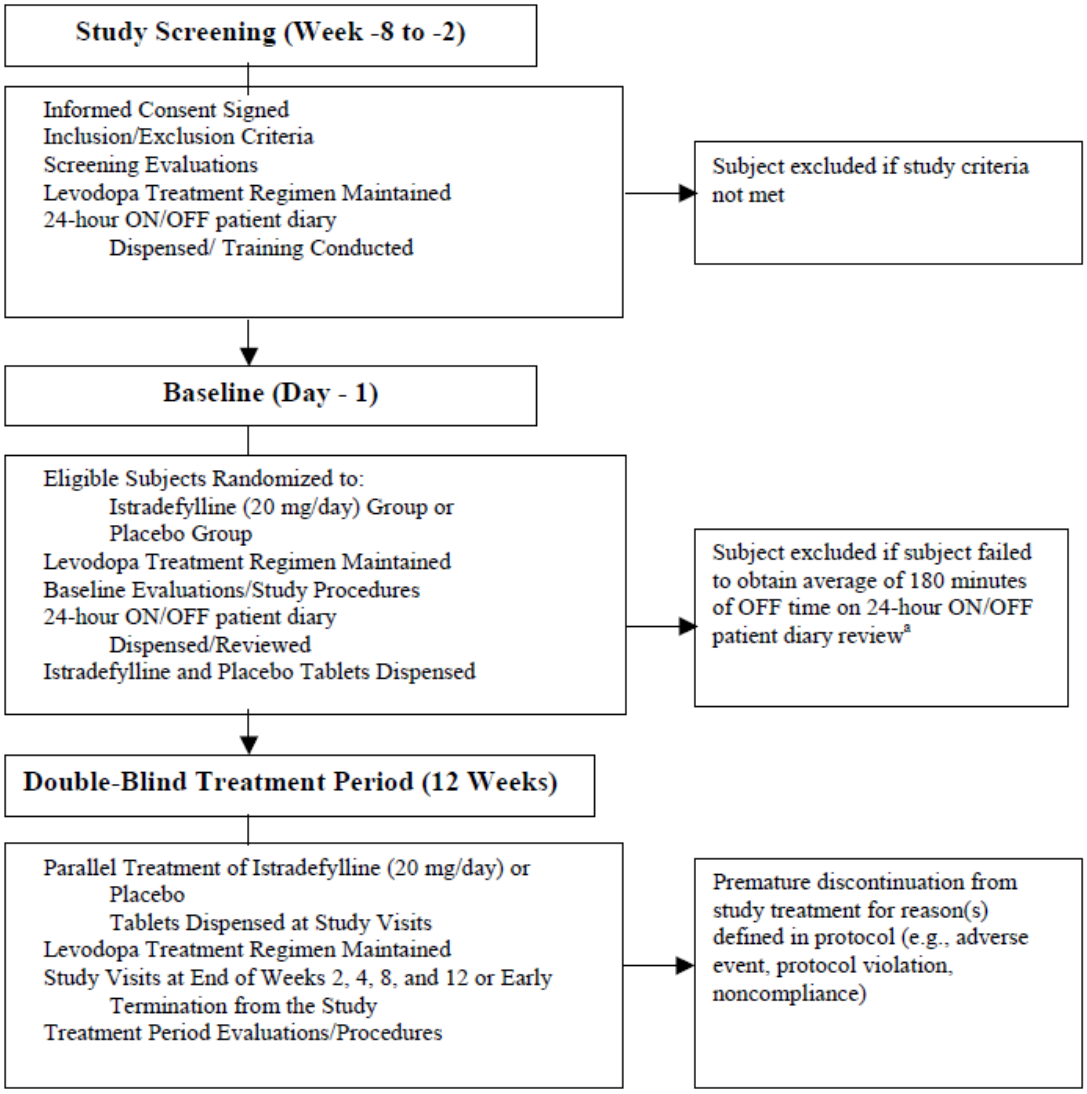
Study 6002-US-013

'A 12-Week, Double-Blind, Placebo-Controlled, Randomized, Parallel-Group, Multicenter, Fixed Dose Study to Evaluate the Efficacy and Safety of a 20 mg/day Oral Dose of KW-6002 (Istradefylline) as Treatment for Parkinson's Disease in Patients with Motor Response Complications on Levodopa/Carbidopa Therapy'

Methods

Study design

Figure 28: Study plan of Study 6002-US-013



a Any subject with > 4 invalid entries in any 1 of the 2 daily 24-hour ON/OFF PD patient diaries was to repeat the baseline (Day-1) visit 7 days later. A subject who failed the validity criterion a second time was excluded from participating in the study.

Study Participants

Inclusion Criteria

- Subjects who met the UKPDS brain bank diagnostic criteria (Step 1 and 2) for Parkinson's disease.
- Subjects who had PD in Stages 2-4 while in the OFF state on the modified H&Y scale
- Subjects who had been on levodopa for at least 1 year, and had been on a stable PD regimen within the normal therapeutic limits including levodopa for at least 4 weeks before Baseline.
- Subjects who currently took at least 3 doses of levodopa per day.
- Subjects who had predictable end-of-dose wearing-off.
- Subjects and study site staff each had to complete a practice 24-hour ON/OFF patient diary and achieve concordance of 80% with respect to both ON and OFF periods at the Week -1 visit.
- Subjects who successfully completed two valid 24-hour ON/OFF patient diaries on any 2 consecutive days during the week (7-day period) before the visit at Baseline.
- Subjects who had an average of at least 180 minutes of OFF time per day on the two 24-hour

ON/OFF patient diaries prior to the Baseline visit.

- Subjects who were male or female and at least 30 years of age.
- Subjects who were female had to be non-pregnant and non-nursing. Women of Child-Bearing Potential had to use a reliable method of contraception (e.g., oral contraceptive or long-term injectable or implantable hormonal contraceptive, double-barrier methods, such as condom and diaphragm, condom and foam, condom and sponge or intra-uterine devices) and have a negative serum pregnancy test at Screening and at Baseline. Women were considered not to be of child-bearing potential if they had been surgically sterilized (physician-documented hysterectomy or tubal ligation) or if they were post-menopausal (complete absence of menses for 2 consecutive years) and a serum FSH level of > 30 IU/L in the absence of hormone replacement therapy.
- Subjects who were willing and able to give written informed consent.

Exclusion Criteria

- Subjects who were currently being treated with any restricted medications listed in the protocol.
- Subjects who were treated within 30 days before Baseline (or 5 half-lives of the compound, if longer) with any investigational agent.
- Subjects who had a history of psychotic illness.
- Subjects who were treated with an anti-psychotic agent within 3 months (6 months if the subject was treated with depot anti-psychotic agent) before Baseline or during the study.
- Subjects who were treated with any centrally-acting drug that has known dopamine antagonist properties at therapeutic doses (e.g., buspirone, amoxapine).
- Subjects who had undergone a neurosurgical operation for PD (e.g., pallidotomy, thalamotomy, deep brain stimulation);
- Subjects who were previously treated with istradefylline.
- Subjects who had atypical parkinsonism.
- Subjects who had secondary parkinsonism variants.
- Subjects who had a diagnosis of cancer or had evidence of continued malignancy within 5 years of study enrollment (except for subjects that have had basal cell carcinoma or carcinoma in situ of the cervix surgically excised).
- Subjects who had a clinically significant illness of any organ system which may have compromised the safety of the Subject during the trial or have affected the ability of the Subject to complete the trial.
- Subjects who, for any reason, were judged by the Investigator to be inappropriate for the study, including a subject who was unable to communicate or to cooperate with the Investigator;
- Subjects who had an ALT and/or an AST level greater than 1.5 times the ULN at Screening were ineligible to participate in the study;
- Subjects with an MMSE score of 25 or less.
- Subjects who had a history of drug or alcohol abuse or dependence within the year prior to enrollment (DSM-IVR).
- Subjects with significant drug allergies.
- Subjects who, in the opinion of the Investigator, had a concurrent clinically relevant depression (whether or not under active treatment).
- Subjects who had a history of seizures or seizure disorders.
- Subjects who had a history of neuroleptic malignant syndrome.

Treatments

The study treatment consisted of one 20 mg istradefylline tablet or one matching placebo tablet once daily.

During the study, subjects were to take levodopa according to the Investigator's clinical judgment and the requirements of the protocol. In addition, other antiparkinson's medications were permitted.

All istradefylline and placebo tablets were supplied by KPI.

Levodopa and any other antiparkinson's medications were to be obtained from local pharmacies.

Prior and concomitant therapy

Investigators were to exercise caution when co-administering medications possibly metabolized by CYP3A4.

Medications prohibited during the study included the following:

- Apomorphine (Screening through Week 12 or at time of early termination from the study);
- Dopamine receptor antagonists (e.g. phenothiazines, clozapine) unless approved by the medical monitor on a per-subject basis; and
- Any investigational agent within 30 days before Baseline (or 5 half-lives of the compound, if longer).

During Screening, subjects were to maintain treatment regimens of levodopa and any other antiparkinson's medications that were obtained from local pharmacies

Levodopa Adjustment and adjustment of other Antiparkinson's Medications

The approach was the same as the one described for study 6002-US-005

Objectives

The primary objective of this study was to evaluate the efficacy of 20 mg/day istradefylline for reducing the percentage of awake time per day spent in the OFF state in subjects with PD treated with L/C hereafter to be referred to as levodopa whenever reference is made to levodopa with a peripheral dopa decarboxylase inhibitor such as carbidopa or benserazide, when used as a drug for a subject) who have motor response complications.

The secondary objectives of this study were to evaluate:

- The efficacy of 20 mg/day istradefylline for reducing the total hours of OFF time.
- The change in total hours of ON time and percentage of ON time without dyskinesia, with dyskinesia, with non-troublesome dyskinesia, with troublesome dyskinesia, and without troublesome dyskinesia (the change in total hours of ON time and percentage of ON time without troublesome dyskinesia was added to the SAP prior to the database lock/unblinding).
- The change in UPDRS Motor Examination Subscale III score and in ADL Subscale II score.
- The change in Parkinson's Disease Questionnaire (PDQ-39 [sum of questions 1 to 39] and PDQ-8 [sum of questions 7, 12, 17, 25, 27, 31, 35, and 37]), and Medical Outcomes Study 36-Item Short Form (SF-36);
- The Patient Global Impression - Improvement scale (PGI-I).
- The change in the Clinical Global Impression - Severity of Illness scale (CGI-S).

- The safety of 20 mg/day istradefylline by changes in safety parameters.

Outcomes/endpoints

Primary Efficacy Outcome

The primary efficacy variable was the percentage of awake time per day spent in the OFF state in subjects with Parkinson's disease treated with L/C.

To meet the primary objective in this study, subjects recorded assessments of ON and OFF time every 30 minutes for 24 hours in the 24-hour ON/OFF patient diary on 2 consecutive days prior to the Baseline, Week 2, Week 4, Week 8, and Week 12 study visits (or the last post-Baseline study visit at the time of the subject's discontinuation from the study). For purposes of analysis, the total hours and percentage of awake time per day spent in the OFF state for the visit was calculated as an average of the values for the 2 diaries completed for the visit.

Secondary Efficacy Outcomes

Secondary efficacy outcomes were defined based on the subject´s valid 24-hour ON/OFF patient diary, UPDRS, CGI-S, PDQ, SF-36 and PGI-I.

Sample size

The sample size per group for testing the primary efficacy variable was based on the expected difference in change from Baseline in the percentage of awake time per day spent in the OFF state at Endpoint between the 20 mg/day istradefylline and placebo groups. A sample size of 100 subjects for each group provided 80% power to detect a difference with an effect size of 40% between the 20 mg/day istradefylline group and the placebo group at the two-sided 5% significance level using a t-test. To account for a small percentage of subjects who were not expected to qualify for the ITT analysis set (i.e., less than 5%), 105 subjects were to be randomized to each group for a total of 210 subjects.

Randomisation

A subject who met all entry criteria was randomized on Day -1 to treatment with 20 mg/day istradefylline or matching placebo in a 1:1 ratio.

All subjects were randomized only once. Once a number was assigned, no attempt was made to use that number again.

If a randomization number was assigned to a subject incorrectly, no attempt was made to remedy the error once the study drug had been dispensed. The subject was to continue with the assigned number and the corresponding study drug.

Blinding (masking)

Tablets were packaged in bottles identical in appearance and each containing 20 tablets.

The subject, site personnel, and the Sponsor were blinded to the treatment group to which a subject was assigned. Study monitors and data management were unaware of the contents of the randomization code until after the study was completed, all data were validated, and the database was locked.

The blind could be broken only if immediate knowledge of the study drug was needed to provide optimal clinical treatment to the subject. No unblinding occurred during the conduct of the study.

Statistical methods

The following study populations were specified: safety analysis set and ITT analysis set as defined in the Study 6002-US-005 and a per protocol (PP) analysis set defined as as subjects who were in the ITT analysis set and who completed 12 weeks of the DB treatment period with no major protocol violations (similar to EFF).

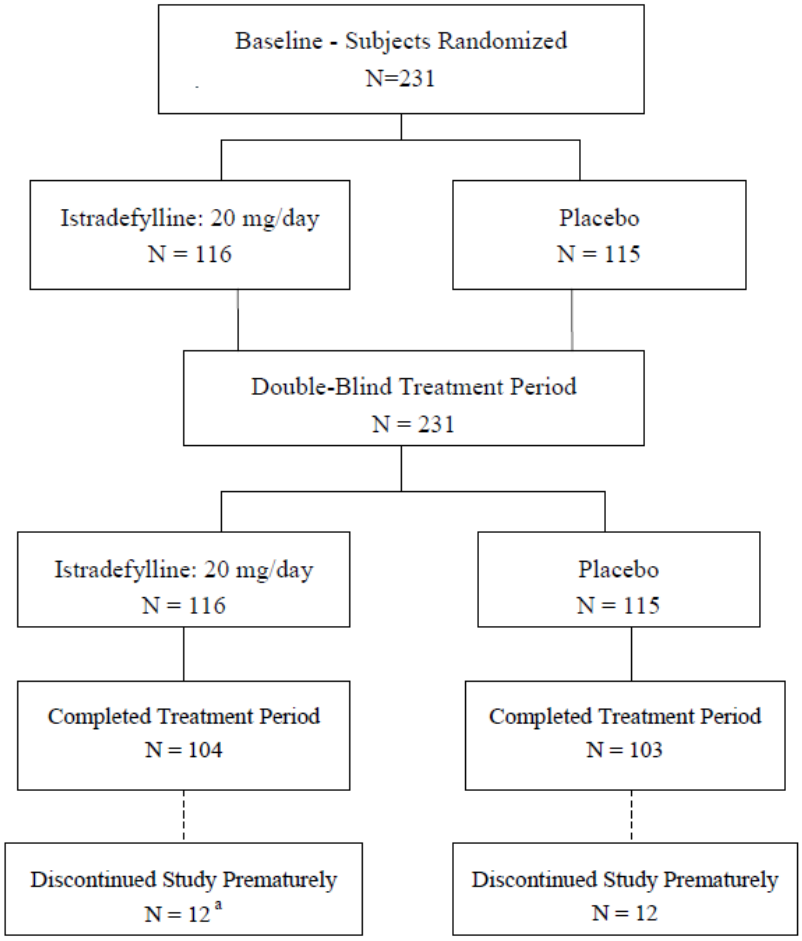
The primary efficacy analyses were based on the ITT analysis set. The PP analysis set was used to carry out a secondary analysis for the primary efficacy variable at Endpoint. The following rules included those protocol violations that were used as the basis to exclude subjects from the PP analysis set.

The main analysis used an ANCOVA model, with use of ANOVA if a significant treatment by baseline interaction was found, and it was qualitative in nature.

Results

Participant flow

Figure 29: Disposition of subjects



a One subject (Subject No.077/101) was randomised to the istradefylline group but did not take study drug and was discontinued from the study. This subject was not included in any of the safety or efficacy analysis.

Recruitment

Study locations: 26 centers in the US

Study period: first dose of study drug was on 14 July 2002 and last dose was 21 November 2005

Conduct of the study

Amendments

The original protocol was issued on 04 May 2004. Three amendments were made to the protocol: Amendment 1 (dated 26 October 2004), Amendment 2 (dated 23 March 2005), and Amendment 3 (dated 29 April 2005).

Amendment 1 included allowing modifying daily dose of study drug and performance of full neurological examination during physical examination. Amendment 2 included revising time period for taking prohibited medications to include 2 weeks following last dose of study drug and amended the list of prohibited medication to allow low doses of antipsychotic medications. Amendment 3 comprised only of deletion of a footnote, to no longer record change in weight of 7lbs or more as an AE on CRF.

Protocol deviations

A total of 28 subjects did not meet all the protocol-specified entry criteria. Six subjects were withdrawn from the study because of protocol violations: 5 subjects in the istradefylline group and 1 subject in the placebo group.

Baseline data

Table 51: Demographic and other baseline characteristics (safety analysis set)

Demographic Characteristic	Placebo N=115	Istradefylline 20 mg/day N=115	Total N=230
Age (years)			
Mean	63.7	63.3	63.5
SD	10.15	9.47	9.79
Median	63.0	63.0	63.0
Range (min-max)	36 to 87	37 to 84	36 to 87
Sex (n [%])			
Male	77 (67.0)	76 (66.1)	153 (66.5)
Female	38 (33.0)	39 (33.9)	77 (33.5)
Race (n [%])			
Caucasian	105 (91.3)	106 (92.2)	211 (91.7)
Black	4 (3.5)	2 (1.7)	6 (2.6)
Asian	5 (4.3)	1 (0.9)	6 (2.6)
Hispanic	1 (0.9)	1 (0.9)	2 (0.9)
American Indian	0	1 (0.9)	1 (0.4)
Other[a]	0	4 (3.5)	4 (1.7)
Height (cm)			
Mean	172.64	172.63	172.63
SD	10.249	8.670	9.472
Median	173.00	173.00	173.00
Range (min-max)	139.7 to 195.6	152.4 to 190.0	139.7 to 195.6
Weight[b] (kg)			
Mean	81.13	80.65	80.89
SD	18.643	18.360	18.464
Median	79.40	78.90	78.90
Range (min-max)	43.3 to 137.1	44.5 to 135.0	43.3 to 137.1
BMI (kg/m^2)			
Mean	27.08	26.92	27.00
SD	5.226	5.139	5.172
Median	26.80	26.30	26.40
Range (min to max)	17.7 to 44.9	17.2 to 41.0	17.2 to 44.9
Current Smoker			
Yes (n [%])	7 (6.1)	4 (3.5)	11 (4.8)
No (n [%])	108 (93.9)	111 (96.5)	219 (95.2)

a "Other" included Iranian, Middle Eastern, Hawaiian (South Pacific Islander), and Italian.
b At screening
SD = standard deviation; min= minimum; max=maximum; cm=centimetre; kg=Kilogram

Table 52: PD history at baseline (safety analysis set)

Characteristic	Placebo N=115	Istradefylline 20 mg/day N=115	Total N=230
Time since diagnosis (years)			
Mean	8.83	10.03	9.43
SD	4.445	5.501	5.026
Median	7.80	8.70	8.05
Range (min to max)	0.1[a] to 24.2	1.3 to 36.8	0.1[a] to 36.8
Time since Initiation of levodopa (years)			
Mean	7.54	8.62	8.08
SD	4.512	5.467	5.030
Median	6.90	8.10	7.50
Range (min to max)	1.2 to 24.2	1.1 to 31.8	1.1 to 31.8
Time since onset of motor response complications (years)			
Mean	3.56	3.99	3.78
SD	3.299	3.723	3.516
Median	2.10	3.00	2.80
Range (min to max)	0.1 to 14.7	0.1 to 24.8	0.1 to 24.8

One subject had an onset of PD value reported as 0.1 years. The same subject began levodopa therapy 3.8 years before and had an onset of motor complications 1.5 years prior to study entry. The time since diagnosis of PD was queried
Duration of PD history was calculated relative to the screening visit date
a Included only subjects who had no missing value for both the month and year of the event time
SD = standard deviation; min= minimum; max=maximum.

Table 53: PD Characteristics at baseline: percentage of awake time per day (safety analysis set)

Characteristic	Placebo N=115	Istradefylline 20 mg/day N=115	Total N=230
In the OFF state[a]			
n[b]	115	113	228
Mean	38.63	39.56	39.09
SD	11.577	14.220	12.934
Median	38.20	38.00	38.10
Range (min to max)	17.6 to 64.7	11.4 to 84.2	11.4 to 84.2
In the ON state without dyskinesia[a]			
n[b]	115	113	228
Mean	48.44	43.56	46.02
SD	18.165	20.166	19.296
Median	50.70	47.00	48.25
Range (min to max)	0.0 to 81.8	0.0 to 81.8	0.0 to 81.8
In the ON state with dyskinesia[a]			
n[b]	115	113	228
Mean	12.93	16.88	14.89
SD	18.482	21.213	19.937
Median	0.00	6.80	3.50
Range (min to max)	0.0 to 73.6	0.0 to 71.0	0.0 to 73.6
In the ON state with non-troublesome dyskinesia[a]			
n[b]	115	113	228
Mean	10.10	12.54	11.31
SD	14.901	16.167	15.555
Median	0.00	4.80	1.45
Range (min to max)	0.0 to 63.9	0.0 to 68.1	0.0 to 68.1
In the ON state with troublesome dyskinesia[a]			
n[b]	115	113	228
Mean	2.83	4.34	3.58
SD	6.363	8.879	7.733
Median	0.00	0.00	0.00
Range (min to max)	0.0 to 31.8	0.0 to 42.5	0.0 to 42.5
In the ON state without troublesome dyskinesia[a]			
n[b]	115	113	228
Mean	58.53	56.10	57.33
SD	11.993	14.405	13.270
Median	58.80	57.60	57.85
Range (min to max)	31.0 to 81.8	15.8 to 82.6	15.8 to 82.6

a Based on valid 24-hour ON/OFF PD patient diaries from observed-case analysis.
b Number of subjects who have available data at baseline.
SD = standard deviation; min= minimum; max=maximum.

Numbers analysed

Table 54: Subject disposition

Status	Placebo N=115 n (%)	Istradefylline 20 mg/day N=116 n (%)
All Subjects Randomized	115 (100.0)	116 (100.0)[a]
Safety Analysis Set[b]	115 (100.0)	115 (99.1)
Intent-to-Treat Analysis Set[c]	113 (98.3)	112 (96.6)
Per-Protocol Analysis Set[d]	99 (86.1)	100 (86.2)
Completed		
Week 2	111 (96.5)	113 (97.4)
Week 4	107 (93.0)	110 (94.8)
Week 8	103 (89.6)	109 (94.0)
Completed 12-Week Double-blind Treatment Period	103 (89.6)	104 (89.7)
Discontinued Study Prematurely	12 (10.4)	12 (10.3)
Reason for Discontinuation from Study		
Lack of efficacy	1 (0.9)	0
Adverse events	7 (6.1)	6 (5.2)
Protocol violation	1 (0.9)	5 (4.3)
Subject withdrew consent	3 (2.6)	0
Other[e]	0	1 (0.9)

a One subject was randomised to the istradefylline group but did not take any study drug and was discontinued from the study. This subject was not included in any efficacy or safety analyses.
b All randomised subjects who received at least 1 dose of study drug
c All randomised subjects who received at least 1 dose of study drug and had a baseline and at least 1 post-baseline 24-hour ON/OFF PD patient diary.
d Subjects who are in the ITT analysis set and who complete 12 weeks of treatment with no major protocol deviations.
e Reason for other was "missed scheduled visits"

Outcomes and estimation

Primary Efficacy Variable

The primary efficacy variable was the change from Baseline in the percentage of awake time per day spent in the OFF state at Endpoint based on the 24-hour ON/OFF patient diary data.

Table 55: Change from baseline at endpoint in percentage of awake time per day spent in an off state (Observed Case Analysis)

	Percentage of Awake Time Per Day Spent in the OFF State[a]		
Visit	Placebo N=113	Istradefylline 20 mg/day N=112	p-value[b]
Baseline			
n	113	112	
Mean	38.72	39.81	
SD	11.612	14.029	
Median	38.20	38.10	
Min to max	17.6 to 64.7	14.6 to 84.2	
Endpoint			
n	113	112	
Mean	33.74	30.48	
SD	17.416	16.315	
Median	32.80	26.70	
Min to max	0.0 to 91.9	0 to 72.4	
Change from Baseline at Endpoint			
n	113	112	
Mean	-4.97	-9.34	
SD	15.468	15.909	
Median	-4.30	-8.85	
Min to max	-58.4 to 43.6	-59.5 to 29.5	
LS mean	-4.92	-9.49	0.025
95% CI (of change)	(-7.76, -2.08)	(-12.43, -6.56)	
LS mean difference (versus placebo)		-4.57	
95% CI (versus placebo)		(-8.55, -0.59)	

a Based on valid 24-hour ON/OFF PD diaries from observed-case analysis
b The p-value is based on Type III Sums of squares F-test with one degree of freedom from the main effects ANCOVA with terms for baseline, investigator and treatment.
CI= confidence interval LSM = Least squares mean; SD = Standard deviation; min= minimum; max=maximum.

Table 56: Sensitivity Analyses of the Primary Endpoint

Analysis Method		Statistic	Placebo	Istradefylline 20 mg/day
WORST CASE				
ANCOVA	WC	Baseline (mean)	38.96%	40.08%
		Endpoint (mean)	34.01%	30.95%
		LS mean change from baseline to endpoint	-4.89%	-9.26%
		LS mean difference from placebo (95% CI)	-	-4.37% (-8.31, -0.43)
HOURS PER DAY				
ANCOVA	OC	Baseline (mean)	6.53 h	6.71 h
		Endpoint (mean)	5.65 h	5.12 h
		LS mean change from baseline to endpoint	-0.86 h	-1.58 h
		LS mean difference from placebo (95% CI)	-	-0.73 h (-1.39, -0.06)
	WC	Baseline (mean)	6.62 h	6.83 h
		Endpoint (mean)	5.74 h	5.26 h
		LS mean change from baseline to endpoint	-0.86 h	-1.55 h
		LS mean difference from placebo (95% CI)	-	-0.69 h (-1.36, -0.03)
SCE ANALYSIS				
MMRM	OC	Baseline (mean)	6.53 h	6.71 h
		Week 12 (mean)	5.43 h	5.20 h
		LS mean change from baseline to Week 12	-0.92 h	-1.53 h
		LS mean difference from placebo (95% CI)	-	-0.62 h (-1.31, -0.08)
	WC	Baseline (mean)	6.53 h	6.83 h
		Week 12 (mean)	5.43 h	5.34 h
		LS mean change from baseline to Week 12	-0.94 h	-1.47 h
		LS mean difference from placebo (95% CI)	-	-0.54 h (-1.23, -0.15)

NA=not applicable; ANCOVA=Analysis of covariance; ANOVA=Analysis of variance; CI=Confidence interval; LS= Least squares; MMRM=Mixed-model repeated measures; OC=Observed case; SCE=Summary of clinical efficacy; WC=Worst Case.

Secondary Efficacy variables

Several analysis were provided by the applicant for secondary efficacy variables. Overall, secondary efficacy endpoints were numerically in favour of istradefylline. In terms of LSM mean difference in total hours of awake time per day spent in OFF state, performed as a secondary efficacy endpoint analysis, this showed a reduction of 0.73 hours for istradefylline 20mg group compared to placebo. This approximates to a difference from placebo of 44 minutes. Moreover, the percentage of awake time/day spent ON without troublesome dyskinesia as a relevant secondary endpoint showed changes numerically favouring istradefylline (see below table). Due to the lack of multiplicity control, interpretation of results from secondary efficacy variables was solely descriptive.

Summary of main efficacy results

This summary should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 57: Summary of efficacy for trial 6002-US-013

Title: A 12-Week, Double-Blind, Placebo-Controlled, Randomized, Parallel-Group, Multicenter, Fixed-Dose Study to Evaluate the Efficacy and Safety of a 20 mg/day Oral Dose of KW-6002 (Istradefylline) as Treatment for Parkinson's Disease in Patients with Motor Response Complications on Levodopa/Carbidopa Therapy				
Study identifier	6002-US-013			
Design	Phase 3, randomised, double-blind, placebo-controlled, fixed-dose, parallel-group, multicenter trial			
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:		12 weeks Not applicable Not applicable	
Hypothesis	Superiority			
Treatments groups	Istradefylline 20 mg		Istradefylline 20 mg once daily, 12 weeks, n=116 subjects	
	Placebo		Placebo once daily, 12 weeks, n=115 subjects	
Endpoints and definitions	Primary endpoint	OFF time per day	Change from baseline in the percentage of awake time/day spent in the OFF state	
	Key secondary endpoint	ON time per day without troublesome dyskinesia	Change from baseline in percentage of awake time/day spent ON without troublesome dyskinesia	
Database lock	12 January 2006			
Results and Analysis				
Analysis description	**Primary Analysis**			
Analysis population and time point description	Intent to treat (ITT), week 12/Endpoint LOCF, Observed case, ANCOVA			
Descriptive statistics and estimate variability	Treatment group		Placebo	Istradefylline 20 mg
	Number of subjects analysed at Week 12/Endpoint		113	112
	Change from baseline in the percentage of awake time/day spent in the OFF state; Mean (SD)		-4.97 (15.468)	-9.34 (15.909)
	Range (min to max)		-58.4 to 43.6	-59.5 to 29.5
	Change from baseline in percentage of awake time/day spent in the ON state without troublesome dyskinesia; Mean (SD)		3.95 (17.020)	7.83 (15.858)
	Range (min to max)		-54.4 to 58.4	-29.5 to 59.5
	Primary endpoint (% of awake time/day spent in the OFF state);		Comparison groups	Istradefylline vs Placebo

Effect estimate per comparison	difference from placebo in the LSM change from baseline	LSM change vs. placebo	-4.57
		95% CI	-8.55, -0.59
		P-value	0.025
	<u>Key secondary endpoint</u> (% of awake time/day spent ON without troublesome dyskinesia); difference from placebo in the LSM change from baseline	Comparison groups	Istradefylline vs Placebo
		LSM change vs. placebo	3.87
		95% CI	-0.35, 8.09

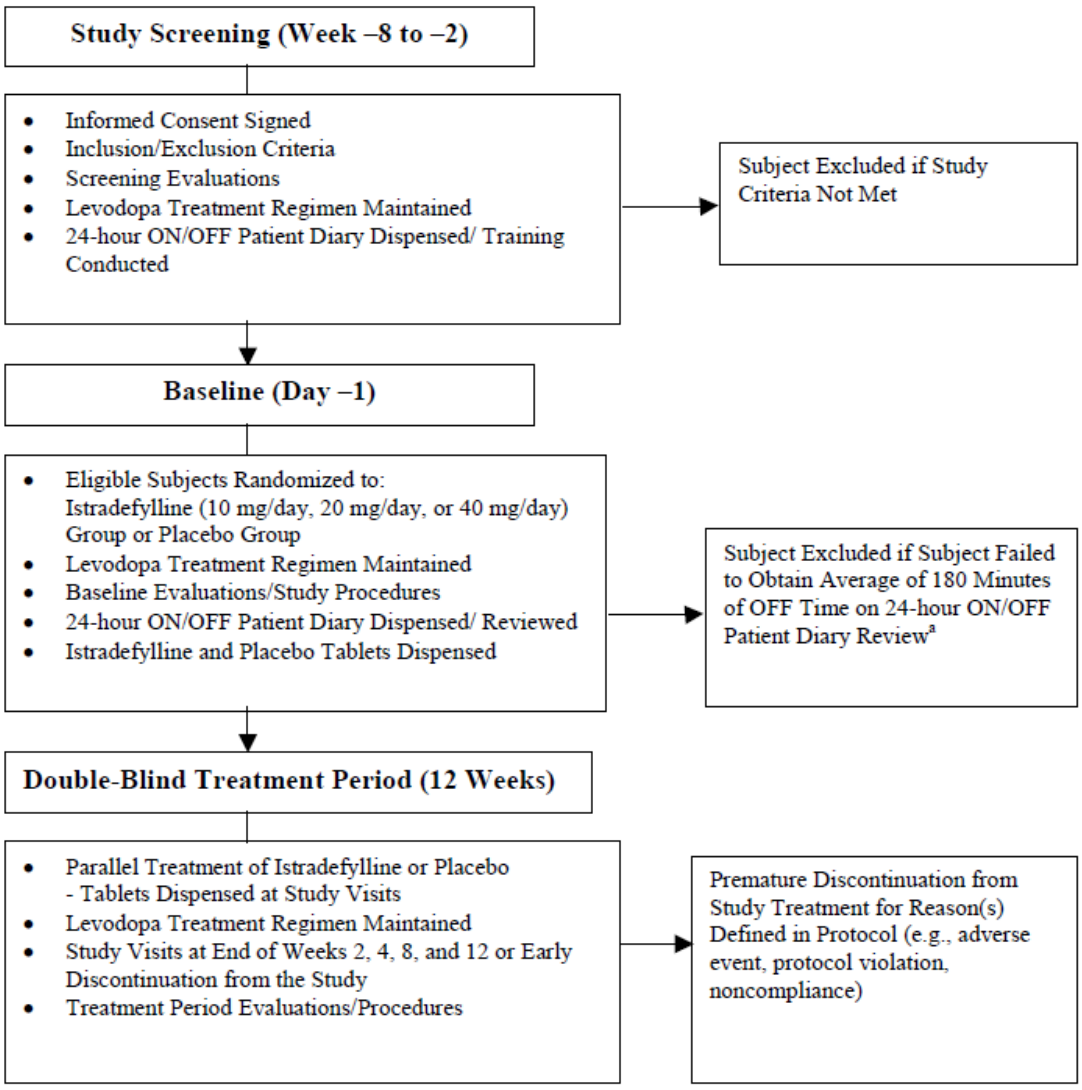
6002-US-018

"A 12-Week, Double-Blind, Placebo-Controlled, Randomized, Parallel-Group, Multicenter, Fixed Dose-Response Study to Evaluate the Efficacy and Safety of 10, 20, and 40 mg/day Oral Doses of KW-6002 (Istradefylline) as Treatment for Parkinson's Disease in Patients with Motor Response Complications on Levodopa/Carbidopa Therapy"

Methods

Study design

Figure 30: Study plan of Study 6002-US-018



a Any subject with > 4 invalid entries in any 1 of the 2 24-hour ON/OFF PD diaries was to repeat the Baseline (Day-1) visit 7 days later. A subject who failed the study entry criteria for diary validation a second time was excluded from participating in the study.

Study Participants

Inclusion and exclusion criteria were the same as for the Study 6002-US-013

Treatments

The study treatment consisted of 10 mg istradefylline tablets, 20 mg istradefylline tablets, 40 mg istradefylline tablets, or matching placebo tablets once daily.

Subjects took 2 tablets each day as follows:

- Subjects in the 10 mg/day istradefylline group took one 10 mg istradefylline tablet plus one matching 40 mg placebo tablet.
- Subjects in the 20 mg/day istradefylline group took one 20 mg istradefylline tablet plus one matching 40 mg placebo tablet.
- Subjects in the 40 mg/day istradefylline group took one 40 mg istradefylline tablet plus one matching 20 mg placebo tablet.

- Subjects in the placebo group took 2 matching placebo tablets (one 20 mg placebo tablet and one 40 mg placebo tablet).

During the study, subjects were to take levodopa according to the Investigator's clinical judgment and the requirements of the protocol. In addition, other antiparkinson's medications were permitted.

Objectives

Primary Objective

The primary objective of this study was to establish the efficacy of 10, 20, and 40 mg/day istradefylline for reducing the percentage of awake time per day spent in the OFF state in subjects with PD treated with L/C who had motor response complications.

Secondary Objectives

The secondary objectives of this study were to evaluate:

- The dose response of 10, 20, and 40 mg/day istradefylline using the primary efficacy outcome variable.
- The efficacy of 10, 20, and 40 mg/day istradefylline for reducing the total hours of OFF time.
- The change in total hours of ON time (without dyskinesia, with dyskinesia, with non-troublesome dyskinesia, and with troublesome dyskinesia).
- The change in UPDRS Motor Examination score (Subscale III) and in ADL score (Subscale II).
- The change in PDQ-39 and SF-36.
- The PGI-I scale.
- The change in CGI-S scale.
- The safety of 10, 20, and 40 mg/day istradefylline by changes in safety parameters.

In addition, the following secondary efficacy objectives were not specified in the protocol but were defined in the SAP prior to the database lock/unblinding.

- The change in PDQ-8 [sum of questions 7, 12, 17, 25, 27, 31, 35, and 37]).
- The change in total hours of ON time without troublesome dyskinesia.
- The change in percentage of ON time (without dyskinesia, with dyskinesia, with non-troublesome dyskinesia, with troublesome dyskinesia, and without troublesome dyskinesia).

Outcomes/endpoints

Primary Efficacy Measurements and Endpoints

The primary efficacy variable of this study was the change from baseline in the percentage of awake time per day spent in the OFF state at Endpoint (Week 12 value or the last available post-Baseline value at the time of premature discontinuation from the study) based on data from the subject's 24-hour ON/OFF patient diary.

Secondary Efficacy Measurements and Endpoints

Several secondary efficacy measurements and endpoints were defined based on the subject´s valid 24-hour ON/OFF patient diary, UPDRS, CGI-S, PDQ, SF-36 and PGI-I.

Sample size

The sample size per treatment group for testing the primary efficacy variable was based on the expected difference in change from Baseline in the percentage of awake time per day spent in the OFF state at Endpoint between the istradefylline groups and placebo. Two recently completed studies evaluating 20, 40, and/or 60 mg/day of istradefylline with placebo (40 mg/day in Study 6002-US-005 and 20 and 60 mg/day in Study 6002-US-006) were used to estimate the expected effect size for determining sample size requirements for this study. The noncentrality parameter for the power calculations was based on estimated means and mean squared errors from this combined analysis.

A sample size of 137 subjects for each treatment group provided 90% power to detect an overall treatment effect at the two-sided 5% significance level using an F-test with two degrees of freedom involving the placebo, 20 mg, and 40 mg groups. Based on the pairwise differences between the 20 mg and 40 mg groups with placebo, standardized effect sizes of 40% or more for one of the treatment groups and 30% or more for the other treatment group satisfies the alternative with the above specifications. These pairwise effect sizes correspond to approximately a 1-hour reduction in OFF time and a 40-minute reduction in OFF time, respectively. To account for a small percentage of subjects who were not expected to qualify for the ITT analysis set (e.g., 2%), 140 subjects were to be randomized to each treatment group for a total of 560 subjects.

Randomisation

At the Baseline visit (Day -1) eligible subjects were randomized in a 1:1:1:1 ratio to treatment with 10 mg/day istradefylline, 20 mg/day istradefylline, 40 mg/day istradefylline, or matching placebo.

Blinding (masking)

The subject, site personnel, and the Sponsor were blinded to the treatment group to which a subject was assigned. Study monitors and data management personnel were unaware of the contents of the randomization code until after the study was completed, all data were validated, and the database was locked. The 10 mg and 20mg istradefylline tablets were identical in appearance. The blind could be only if immediate knowledge of the study drug was needed to provide optimal clinical treatment to the subject. No unblinding occurred during the conduct of the study.

Statistical methods

The SAP for this study was developed and finalized prior to the unblinding of treatment codes. The final SAP was signed on 04 November 2005 with an erratum issued on 22 November 2005. The database was locked on 03 January 2006 and data were unblinded for the efficacy analyses on 04 January 2006.

The following analysis sets of the study populations were used: safety analysis set, ITT analysis set and PP analysis set as described for Study 6002-US-13.

All safety analyses were carried out using the safety analysis set. The primary efficacy analyses were based on the ITT analysis set. The PP analysis set was used to carry out a secondary analysis for the primary efficacy variable at Endpoint

Results

Participant flow

Table 58: Subject Disposition

Status	Placebo N=154 n (%)	Istradefylline 10 mg/day N=155 n (%)	Istradefylline 20 mg/day N=149 n (%)	Istradefylline 40 mg/day N=152 n (%)	Total N=610 n (%)
All Subjects Randomized	154 (100)	155 (100)	149 (100)	152 (100)	610 (100)
Safety Analysis Set[a]	151 (98.1)	153 (98.7)	149 (100)	152 (100)	605 (99.2)[b]
Intent-to-Treat Analysis Set[c]	146 (94.8)	149 (96.1)	144 (96.6)	145 (95.4)	584 (95.7)
Per-Protocol Analysis Set[d]	130 (84.4)	122 (78.7)	117 (78.5)	117 (77.0)	486 (79.7)
Completed					
Week 2	146 (94.8)	148 (95.5)	148 (99.3)	147 (96.7)	589 (96.6)
Week 4	142 (92.2)	146 (94.2)	141 (94.6)	142 (93.4)	571 (93.6)
Week 8	140 (90.9)	138 (89.0)	134 (89.9)	138 (90.8)	550 (90.2)
Completed 12-Week Double-Blind Treatment Period	140 (90.9)	136 (87.7)	131 (87.9)	135 (88.8)	542 (88.9)
Discontinued prematurely	14 (9.1)	19 (12.3)	18 (12.1)	17 (11.2)	68 (11.1)
Reason for Failure to Complete Double-Blind Treatment Period:					
Lack of efficacy	1 (0.6)	2 (1.3)	1 (0.7)	0	4 (0.7)
Adverse events	7 (4.5)[e]	6 (3.9)[f]	15 (10.1)[g]	15 (9.9)[h]	43 (7.0)
Protocol violation	1 (0.6)	4 (2.6)	1 (0.7)[g]	0	6 (1.0)
Subject withdrew consent	3 (1.9)	5 (3.2)[f]	1 (0.7)	1 (0.7)	10 (1.6)
Other	2 (1.3)	2 (1.3)	0	1 (0.7)	5 (0.8)

a All randomized subjects who took at least 1 dose of double-blind study drug.
b Five randomized subjects did not receive study drug (one subject randomized to the placebo group withdrew consent, a different subject randomized to the placebo group was diagnosed with testicular cancer (protocol violation) on the day of randomization and was withdrawn from the study; a third subject randomized to the placebo group and two subjects randomization to the 10 mg/day istradefylline group were lost to follow-up.
c All randomized subjects who took at least 1 dose of double-blind study drug and had a valid Baseline and at least 1 valid post-Baseline 24-hour ON/OFF patient diary.
d Subjects who are in the ITT analysis set and who complete 12 weeks of treatment with no major protocol violations.
e Six subjects were discontinued from the study because of TEAEs and one subject was discontinued from the study because of an AE (liver function test abnormal) that began prior to the first dose of study medication.
f One subject who was discontinued from the study because of an SAE. was inadvertently counted among subjects who withdrew consent. In this in-text table, this subject is included in the counts of subjects who discontinued from the study because of a TEAE, and is not included in the counts of subjects who withdrew consent.
g One subject who was discontinued from the study because of a TEAE (blood creatine phosphokinase increased) was inadvertently counted among the subjects who were discontinued because of a protocol violation. In this in-text table, this subject is included in the counts of subjects who discontinued from the study because of a TEAE, and is not included in the counts of subjects who discontinued from the study because of a protocol violation.
h One subject who was discontinued because of an adverse event (arthralgia) that began 14 days after the last dose of study medication; this subject is not included in the counts of subjects who discontinued because of a TEAE

Recruitment

Study locations: 59 centers in the US and 15 centers in Canada.

Study period: first dose of study drug was on 23 July 2004 and last dose was 16 November 2005.

Conduct of the study

Protocol amendments

The original protocol was issued on 4 May 2004. Three amendments were made to the protocol: Amendment 1 (26 October 2004); Amendment 2 (23 March 2005); and Amendment 3 (29 April 2005). Changes made in these amendments were similar to those of Study US-013 respective amendments and overall no major changes were introduced.

Protocol Deviation

A total of 39 subjects (6.4%) did not meet all protocol-specified entry criteria. Seven subjects, in the istradefylline 10 mg/day (4) and 20 mg/day (2) and placebo (1) groups, were withdrawn from the study because of protocol violations

Baseline data

Table 59: Demographic and other baseline characteristics (safety analysis set)

Demographic Characteristic	Placebo N=151	Istradefylline 10 mg/day N=153	Istradefylline 20 mg/day N=149	Istradefylline 40 mg/day N=152	Total N=605
Age (years)					
n	151	153	149	152	605
Mean	62.7	63.2	63.9	62.9	63.2
SD	8.26	8.89	9.81	9.26	9.06
Median	63.0	64.0	64.0	62.0	63.0
Range (min to max)	41 to 87	43 to 84	35 to 84	35 to 84	35 to 87
Sex (n [%])					
Male	97 (64.2)	103 (67.3)	103 (69.1)	100 (65.8)	403 (66.6)
Female	54 (35.8)	50 (32.7)	46 (30.9)	52 (34.2)	202 (33.4)
Race/ethnic origin (n [%])					
Caucasian	136 (90.1)	142 (92.8)	143 (96.0)	141 (92.8)	562 (92.9)
Black	0	3 (2.0)	2 (1.3)	2 (1.3)	7 (1.2)
Asian	3 (2.0)	3 (2.0)	1 (0.7)	4 (2.6)	11 (1.8)
Hispanic	8 (5.3)	3 (2.0)	2 (1.3)	4 (2.6)	17 (2.8)
American Indian	1 (0.7)	0	0	0	1 (0.2)
Other[a]	3 (2.0)	2 (1.3)	1 (0.7)	1 (0.7)	7 (1.2)
Height (cm)					
n	150	153	149	152	604
Mean	169.45	171.10	171.73	170.04	170.58
SD	10.558	9.450	9.532	10.777	10.111
Median	170.20	171.00	171.00	170.20	170.20
Range (min to max)	142.2 to 190.5	143.0 to 193.0	151.0 to 193.0	147.3 to 198.1	142.2 to 198.1
Weight (kg)[b]					
n	149	153	149	152	603
Mean	80.32	83.57	82.36	81.05	81.83
SD	18.300	16.509	17.513	18.551	17.732
Median	80.50	83.80	81.60	79.85	81.60
Range (min to max)	39.8 to 136.1	44.9 to 132.5	46.7 to 139.3	45.4 to 150.1	39.8 to 150.1
BMI (kg/m^2)					
n	149	153	149	152	603
Mean	27.76	28.51	27.86	28.01	28.04
SD	4.999	5.243	5.423	5.963	5.415
Median	27.00	28.00	27.10	27.25	27.40
Range (min to max)	15.7 to 46.9	18.5 to 47.4	17.8 to 54.4	17.5 to 45.2	15.7 to 54.4
Current Smoker					
Yes	9 (6.0)	3 (2.0)	9 (6.0)	6 (3.9)	27 (4.5)
No	142 (94.0)	150 (98.0)	140 (94.0)	146 (96.1)	578 (95.5)

a "Other" included East Indian, Eastern-European, European White, Indian, Japanese/Portuguese and Portuguese.
b At screening
SD = standard deviation; min= minimum; max=maximum; cm=centimetre; kg=Kilogram

Table 60: PD history at baseline (safety analysis set)

Characteristic	Placebo N=151	Istradefylline 10 mg/day N=153	Istradefylline 20 mg/day N=149	Istradefylline 40 mg/day N=152	Total N=605
Time since Diagnosis (years)					
n[a]	151	153	149	152	605
Mean	9.14	9.11	8.85	8.47	8.89
SD	5.097	4.745	4.631	4.599	4.768
Median	7.70	8.20	7.90	7.40	7.90
Range (min to max)	1.1 to 27.9	1.0 to 30.4	1.0 to 28.2	1.1 to 22.2	1.0 to 30.4
Time Since Initiation of Levodopa (years)					
n[a]	151	153	149	152	605
Mean	7.77	7.89	7.36	7.18	7.55
SD	5.125	4.729	4.411	4.784	4.767
Median	6.90	6.90	6.50	6.05	6.70
Range (min to max)	1.0 to 25.8	1.0 to 30.4	1.0 to 24.8	0.4 to 20.9	0.4 to 30.4
Time Since Onset of Motor Complications (years)					
n[a]	151	153	148	151	603
Mean	3.73	3.63	3.70	3.40	3.61
SD	3.540	3.656	3.383	3.235	3.452
Median	2.50	2.50	2.90	2.30	2.50
Range (min to max)	0.1 to 17.2	0.1 to 24.4	0.0 to 18.3	0.1 to 18.1	0.0 to 24.4

Duration of PD history was calculated relative to the screening visit date
a Included only subjects who had no missing value for both the month and year of the event time
SD = standard deviation; min= minimum; max=maximum.

Table 61: PD Characteristics at baseline: percentage of awake time per day (safety analysis set)

	Percentage of Awake Time per Day				
Characteristic	Placebo N=151	Istradefylline 10 mg/day N=153	Istradefylline 20 mg/day N=149	Istradefylline 40 mg/day N=152	Total N=605
In the OFF state[a]					
n[b]	148	152	146	150	596
Mean	39.77	39.72	39.65	41.70	40.21
SD	11.419	11.884	12.439	13.400	12.308
Median	38.50	39.10	38.15	40.85	39.35
Range (min to max)	11.4 to 73.2	12.9 to 72.3	17.1 to 74.8	12.9 to 79.7	11.4 to 79.7
In the ON state without dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	45.31	45.04	47.53	45.67	45.88
SD	17.842	18.571	18.022	19.955	18.600
Median	46.30	46.80	50.40	50.10	48.20
Range (min to max)	0.0 to 79.7	0.0 to 82.4	0.0 to 82.9	0.0 to 77.1	0.0 to 82.9
In the ON state with dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	14.93	15.24	12.83	12.62	13.91
SD	18.037	19.854	19.386	18.056	18.843
Median	7.30	2.90	0.00	0.00	1.60
Range (min to max)	0.0 to 63.2	0.0 to 73.3	0.0 to 76.4	0.0 to 72.9	0.0 to 76.4
In the ON state with non-troublesome dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	11.52	11.89	10.05	10.63	11.03
SD	14.188	16.020	15.900	16.228	15.587
Median	5.85	2.85	0.00	0.00	1.40
Range (min to max)	0.0 to 58.8	0.0 to 73.3	0.0 to 76.4	0.0 to 72.9	0.0 to 76.4
In the ON state with troublesome dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	3.41	3.35	2.77	2.00	2.88
SD	7.099	7.399	6.459	6.623	6.915
Median	0.00	0.00	0.00	0.00	0.00
Range (min to max)	0.0 to 31.2	0.0 to 53.4	0.0 to 41.3	0.0 to 45.8	0.0 to 53.4
In the ON state without troublesome dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	56.82	56.93	57.58	56.30	56.91
SD	12.464	12.231	11.805	14.363	12.735
Median	56.65	57.60	58.85	58.10	58.10
Range (min to max)	23.6 to 84.5	6.8 to 82.4	23.7 to 82.9	0.0 to 82.4	0.0 to 84.5

a Based on subjects valid ON/OFF PD diaries from observed-case analysis
b The number of subjects who have available data at baseline
NA = not available; SD = Standard deviation; min= minimum; max=maximum.

Table 62: PD characteristics at baseline: total hours of awake time per day (safety analysis set)

	Total Hours of Awake Time per Day				
Characteristic	Placebo N=151	Istradefylline 10 mg/day N=153	Istradefylline 20 mg/day N=149	Istradefylline 40 mg/day N=152	Total N=605
In the OFF state[a]					
n[b]	148	152	146	150	596
Mean	6.70	6.60	6.68	6.90	6.72
SD	2.109	2.035	2.179	2.336	2.164
Median	6.50	6.50	6.30	6.80	6.50
Range (min to max)	2.0 to 13.3	2.0 to 12.0	3.0 to 12.3	1.8 to 16.3	1.8 to 16.3
In the ON state without dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	7.60	7.47	8.02	7.58	7.66
SD	3.065	3.117	3.043	3.414	3.163
Median	8.00	7.50	8.30	8.00	8.00
Range (min to max)	0.0 to 13.5	0.0 to 14.0	0.0 to 14.5	0.0 to 14.0	0.0 to 14.5
In the ON state with dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	2.49	2.59	2.17	2.08	2.33
SD	3.013	3.340	3.222	2.976	3.141
Median	1.30	0.50	0.00	0.00	0.30
Range (min to max)	0.0 to 11.3	0.0 to 11.3	0.0 to 11.8	0.0 to 12.3	0.0 to 12.3
In the ON state with non-troublesome dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	1.92	2.02	1.70	1.77	1.85
SD	2.355	2.659	2.598	2.722	2.585
Median	0.90	0.50	0.00	0.00	0.30
Range (min to max)	0.0 to 9.8	0.0 to 11.0	0.0 to 11.0	0.0 to 12.3	0.0 to 12.3
In the ON state with troublesome dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	0.58	0.58	0.47	0.32	0.49
SD	1.190	1.301	1.114	1.037	1.167
Median	0.00	0.00	0.00	0.00	0.00
Range (min to max)	0.0 to 5.3	0.0 to 9.8	0.0 to 7.3	0.0 to 7.3	0.0 to 9.8
In the ON state without troublesome dyskinesia[a]					
n[b]	148	152	146	150	596
Mean	9.51	9.47	9.70	9.34	9.51
SD	2.241	2.293	2.130	2.604	2.323
Median	9.80	9.50	9.80	9.50	9.80
Range (min to max)	4.0 to 15.0	1.3 to 15.0	3.5 to 16.0	0.0 to 14.3	0.0 to 16.0

a Based on subjects valid ON/OFF PD diaries from observed-case analysis
b The number of subjects who have available data at baseline
NA = not available; SD = Standard deviation; min= minimum; max=maximum.

Outcomes and estimation

Primary Efficacy Variable

The primary efficacy variable was the change from Baseline to Endpoint in the percentage of awake time per day spent in the OFF state based on the subjects' valid 24-hour ON/OFF patient diary data.

Dose-ordering responses within the istradefylline groups (10 mg, 20 mg, and 40 mg/day, respectively) are noted for the primary and secondary efficacy variables, where applicable

Table 63: Change from baseline at endpoint in percentage of awake time per day spent in an OFF state (observed case analysis)

	Percentage of Awake Time Per Day Spent in the OFF State[a]				
Visit	Placebo N=146	Istradefylline 10 mg/day N=149	Istradefylline 20 mg/day N=144	Istradefylline 40 mg/day N=145	Overall p-value[b]
Baseline					
n	146	149	144	145	
Mean	39.76	39.50	39.66	41.78	
SD	11.484	11.714	12.461	13.127	
Median	38.50	38.90	38.15	40.60	
Min to max	11.4 to 73.2	12.9 to 72.3	17.1 to 74.8	13.2 to 79.7	
Endpoint					
n	146	149	144	145	
Mean	32.18	33.84	33.55	32.70	
SD	15.483	16.999	18.692	18.101	
Median	31.90	33.00	30.85	32.90	
Min to max	0.0 to 62.8	0.0 to 83.1	0.0 to 100.0	0.0 to 100.0	
Change from Baseline at Endpoint					
n	146	149	144	145	
Mean	-7.59	-5.67	-6.11	-9.08	
SD	14.367	14.118	17.619	17.606	
Median	-6.15	-6.20	-6.15	-8.60	
Min to max	-59.9 to 28.6	-40.2 to 37.2	-62.0 to 57.6	-64.6 to 48.2	
LS mean	-8.31	-6.52	-6.81	-8.97	0.475
LS mean difference (versus placebo)		1.79	1.50	-0.66	
p-value (versus placebo)[c]		0.319	0.408	0.714	

a Based on subjects' valid 24-hour ON/ OFF PD patient diary data from observed-case analysis.
b The overall p-value for treatment effects is among placebo, 20 mg/day, and 40 mg/day groups only, based on the Type III sums of squares F-test with 2 degrees of freedom.
c The pairwise p-value (istradefylline groups versus placebo group) based on LSM from main effects ANCOVA with terms for Baseline, Investigator, and treatment. A closed testing procedure was used to test for treatment effects; however, all p-values are displayed in this table for descriptive purposes.
LSM = least squares mean; min = minimum; max = maximum; SD = standard deviation

Subgroup Analyses

There were no pre-planned subgroup analyses. However, subgroup analyses by various factors for the change from Baseline in the percentage of awake time per day spent in the OFF state at Endpoint were defined in the SAP prior to database lock.

Summary of main efficacy results

This summary should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 64: Summary of efficacy for trial 6002-US-018

***Title**: A 12-Week, Double-Blind, Placebo-Controlled, Randomized, Parallel-Group, Multicenter, Fixed Dose-Response Study to Evaluate the Efficacy and Safety of 10, 20, and 40 mg/day Oral Doses of KW-6002 (Istradefylline) as Treatment for Parkinson's Disease in Patients with Motor Response Complications on Levodopa/Carbidopa Therapy*		
Study identifier	*6002-US-018*	
Design	*Phase 3, randomised, double-blind, placebo-controlled, fixed-dose, parallel-group, multicenter trial*	
	Duration of main phase:	*12 weeks*
	Duration of Run-in phase:	*Not applicable*

	Duration of Extension phase:		*Not applicable*
Hypothesis	*Superiority*		
Treatments groups	*Istradefylline 10 mg*		*Istradefylline 10 mg once daily, 12 weeks, n=155 subjects*
	Istradefylline 20 mg		*Istradefylline 20 mg once daily, 12 weeks, n=149 subjects*
	Istradefylline 40 mg		*Istradefylline 40 mg once daily, 12 weeks, n=152 subjects*
	Placebo		*Placebo once daily, 12 weeks, n=154 subjects*
Endpoints and definitions	*Primary endpoint*	*OFF time per day*	*Change from baseline in the percentage of awake time/day spent in the OFF state*
	Key secondary endpoint	*ON time per day without troublesome dyskinesia*	*Change from baseline in percentage of awake time/day spent ON without troublesome dyskinesia*
Database lock	*03 January 2006*		

Results and Analysis

Analysis description	***Primary Analysis***				
Analysis population and time point description	*Intent to treat (ITT), week 12/Endpoint LOCF, Observed case, ANCOVA*				
Descriptive statistics and estimate variability	*Treatment group*	*Placebo*	*Istradefylline 10 mg*	*Istradefylline 20 mg*	*Istradefylline 40 mg*
	Number of subjects analysed at Week 12/ Endpoint	*146*	*149*	*144*	*145*
	Change from baseline in the % of awake time/day spent in the OFF state; Mean (SD)	*-7.59 (14.367)*	*-5.67 (14.118)*	*-6.11 (17.619)*	*-9.08 (17.606)*
	Range (min to max)	*-59.9 to 28.6*	*-40.2 to 37.2*	*-62.0 to 57.6*	*-64.6 to 48.2*
	Change from baseline in % of awake time/day spent in the ON state without troublesome dyskinesia; Mean (SD)	*6.61 (15.754)*	*4.19 (15.243)*	*4.88 (18.182)*	*6.79 (19.615)*
	Range (min to max)	*-33.6 to 59.9*	*-37.2 to 38.5*	*-57.6 to 62.0*	*-65.0 to 62.6*
Effect estimate per comparison	*Primary endpoint (% of awake time/day spent in*	*Comparison groups*	*Istradefylline 10mg vs Placebo*	*Istradefylline 20mg vs Placebo*	*Istradefylline 40mg vs Placebo*

	the OFF state); difference from placebo in the LSM change from baseline	*LSM change vs. placebo*	*1.79*	*1.50*	*-0.66*
		95% CI	*-1.73, 5.31*	*-2.05, 5.05*	*-4.21, 2.88*
		P-value	*0.319*	*0.408*	*0.714*
	<u>Key secondary endpoint</u> (% of awake time/day spent ON without troublesome dyskinesia); difference from placebo in the LSM change from baseline	*Comparison groups*	*Istradefylline 10mg vs Placebo*	*Istradefylline 20mg vs Placebo*	*Istradefylline 40mg vs Placebo*
		LSM change vs. placebo	*-2.40*	*-1.55*	*-0.12*
		95% CI	*-6.18, 1.37*	*-5.36, 2.25*	*-3.91, 3.68*

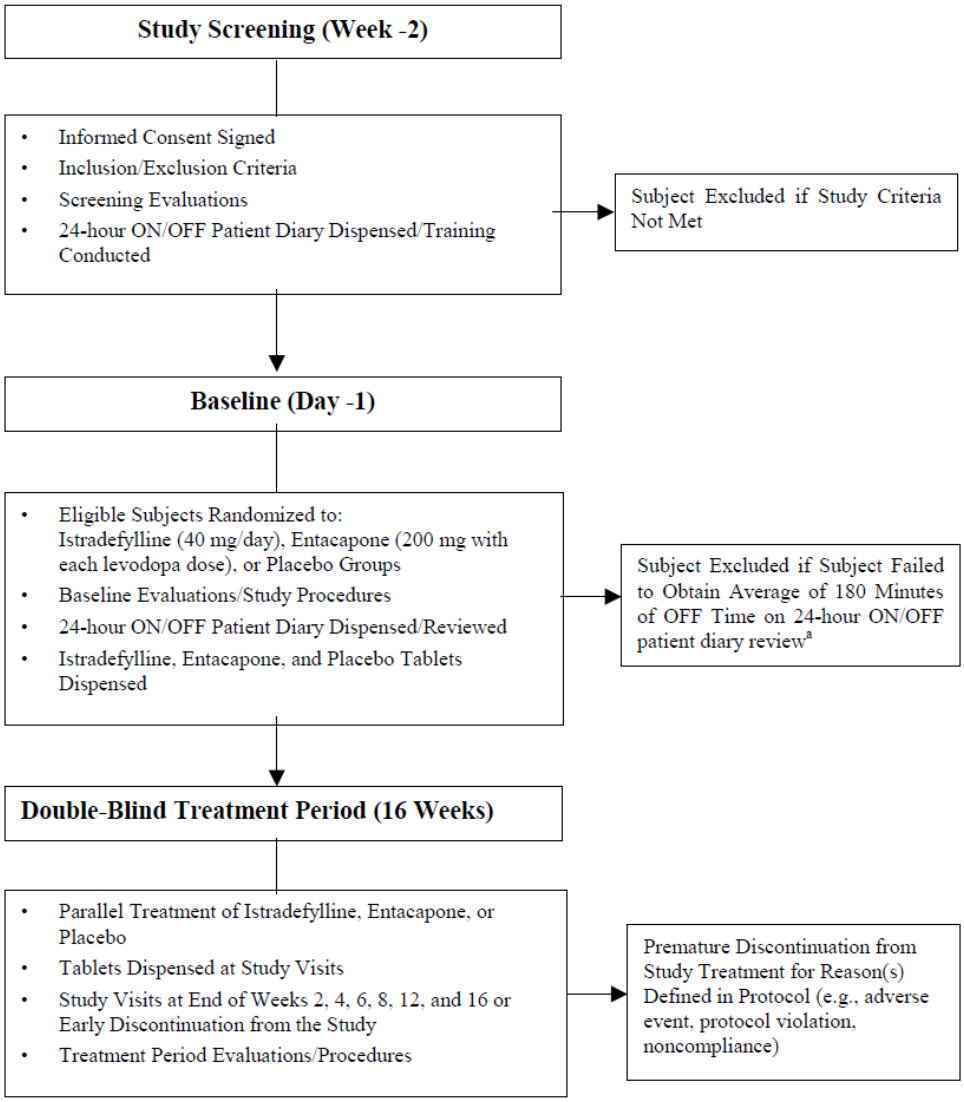
Study 6002-EU-007

"*A 16-week, double blind, placebo-controlled, randomised, parallel group, multicentre, international study to evaluate the efficacy and safety of 40mg/day KW-6002 (istradefylline) and that of entacapone versus placebo as treatment for Parkinson's disease in patients with motor response complications on levodopa therapy*."

Methods

Study design

Figure 31: Study plan of Study 6002-EU-007



a Any subject with > 4 invalid entries in any 1 of the 2 24-hour ON/OFF PD diaries was to repeat the Baseline (Day-1) visit 7 days later. A subject who failed the study entry criteria for diary validation a second time was excluded from participating in the study.

Study participants

Inclusion criteria

- Patients aged 30 or more with PD who met UKPDS brain bank diagnostic criteria (Step 1 and 2).
- PD in in stages 2-4 on modified H&Y scale while in the OFF state. Predictable end of dose wearing off.
- On standard preparations of levodopa with carbidopa or benserazide for at least 1 year; on a stable PD regimen within normal therapeutic limits for at least 4 weeks prior to Baseline; and taking at least 3 doses of levodopa per day.
- Patients and staff must have successfully completed two 'valid' diaries on any 2 consecutive days during the week (7-day period) before the visit at Baseline and patients must have had an average at least 3 hours of OFF time on 2 consecutive diaries prior to the Baseline visit, and not more than 4 invalid entries per 24-hour ON/OFF patient diary.

Main exclusion criteria

- Atypical or secondary parkinsonism
- Prior neurosurgical treatment for Parkinson's
- History of psychotic illness or treated within 3 months (6 months if a depot) before Baseline with an antipsychotic agent apart from subjects experiencing levodopa or Parkinson's disease-induced hallucinations with insight retained where atypical antipsychotic agents may have been allowed at the discretion of the designated Medical Officer (applicable only to subjects entered under Amendment 3);
- Treated within 30 days before baseline with any investigational agent
- Patients currently being treated with restricted medications (Tolcapone (Tasmar®); Apomorphine; Non-selective MAO-A / MAO-B e inhibitors; Dopamine receptor antagonists (e.g. phenothiazines, clozapine) unless approved by the designated Medical Officer on a per-patient basis (this applied to the original protocol and Amendments 1 and 2); Dopamine receptor antagonists (with the exception of atypical antipsychotic treatment of subjects with treatment-emergent levodopa or Parkinson's disease induced hallucinations with insight retained where, with approval of the designated Medical Officer, quetiapine [at a maximum dose of 100 mg/day] or, alternatively, clozapine, [at a maximum dose of 50 mg/day] was recommended) (This applied to subjects entered under Amendment 3. Subjects entered under Amendments 1 and 2 may have been granted a waiver); and controlled or sustained release formulations of levodopa (except for the last dose in a day)
- Previously treated with Istradefylline.

Treatments

Subjects were allocated to 1 of the following randomized treatment groups:

- Group 1: Istradefylline (40 mg) to be taken orally with the first daily dose of levodopa, and placebo orally with each subsequent dose of levodopa.
- Group 2: Entacapone (200 mg) to be taken orally with each dose of levodopa.
- Group 3: Placebo to be taken orally with each dose of levodopa.

Changes to the daily dose or the dose regimen of double-blind medication were not allowed. During the first 4 weeks of the 16-week double-blind treatment period only, adjustment of the levodopa dosage was permitted at the Investigator's discretion if required. However, changes in the interval between levodopa doses were not permitted during this time.

Objectives

The primary objective of this study was to establish the efficacy of 40 mg/day istradefylline in reducing the percentage of awake time per day spent in the OFF state in subjects with PD treated with levodopa.

Note: In this report, levodopa refers to the use of levodopa with a dopa decarboxylase inhibitor such as carbidopa or benserazide, unless otherwise noted.

The secondary objectives of this study were to evaluate:

- The efficacy of a 40 mg/day dose of istradefylline for reducing the total hours of OFF time.
- The change in total hours of ON time and percentage of ON time without dyskinesia, with dyskinesia, with non-troublesome dyskinesia, with troublesome dyskinesia, and without troublesome dyskinesia (the change in total hours of ON time and percentage of ON time without troublesome dyskinesia was added to the SAP prior to the database lock/unblinding).
- The change in UPDRS Subscale I score (Mentation, Behaviour and Mood), Subscale II score (ADL), Subscale III score (Motor Examination), and Subscale IVA score (Dyskinesia).

- The change in the PDQ-39 and PDQ-8 and SF-36 (the PDQ-8 was added to the SAP prior to the database lock/unblinding).
- The PGI-I.
- The change in the CGI-S.
- The safety of a 40 mg/day dose of istradefylline by evaluation of changes in safety parameters.

Outcomes/endpoints

The primary efficacy variable of this study was the change from Baseline in the percentage of awake time per day spent in the OFF state at Endpoint (Week 16 value or the last available post-Baseline value at the time of premature discontinuation from the study) based on data from the 24-hour ON/OFF patient diary.

To meet the primary objective in this study, subjects recorded assessments of ON and OFF time every 30 minutes for 24 hours in the 24-hour ON/OFF patient diary on 2 consecutive days prior to the Baseline, Week 2, Week 4, Week 6, Week 8, Week 12, and Week 16 study visits (or the last post-Baseline study visit at the time of the subject's discontinuation from the study). For purposes of analysis, the total hours and percentage of awake time per day spent in the OFF state for the visit was calculated as an average of the values for the 2 diaries completed for the visit.

Only valid diaries were used in the analysis. A 24-hour ON/OFF patient diary was considered valid if it contained no more than 4 half-hour invalid time points. Invalid time points on a diary were defined as multiple entries (i.e., more than 1 check mark per requested response in a given 30-minute time period) or missing entries (i.e., the absence of an entry in a given 30-minute time period on the diary). Two approaches, an observed-case (primary) and a worst-case (secondary) were used for handling invalid time points on the 24-hour patient diaries. The observed-case analysis treated all invalid time points as missing. The worst-case analysis imputed OFF for missing time points. For time points with multiple entries, the ordering for selecting the worst-case was based on the following: OFF state was the worst-case followed by ON state with troublesome dyskinesia, ON state with non-troublesome dyskinesia, ON state without dyskinesia, and asleep.

Secondary efficacy measurements were based on the changes in the valid 24-hour ON/OFF patient diary data, the UPDRS subscale scores, the CGI-S scale, the PDQ-39, the PDQ-8, the SF-36, and the PGI-I scale.

The secondary efficacy variable of awake time per day spent in the ON state without troublesome dyskinesia was not specified in the protocol, but was defined as the sum of the awake time per day spent in the ON state *without dyskinesia* plus the awake time per day spent in the ON state *with non-troublesome dyskinesia* as a measurement of "functional ON time." This secondary efficacy variable (i.e., ON state without troublesome dyskinesia) was defined in the final SAP, which was completed prior to the database lock and unblinding of this study.

Sample size

The sample size per treatment group for testing the primary efficacy variable was based on the expected difference in change from Baseline in the percentage of awake time per day spent in the OFF state at Endpoint between the 40 mg/day istradefylline treatment group and placebo. A sample size of 130 subjects for each treatment group was estimated to provide 80% power to detect a difference with an effect size of 35% between the 40 mg/day treatment group and the placebo treatment group at the two-sided 5% significance level using a t-test derived from the main effects ANCOVA model. To account for

a small percentage of subjects who were not expected to qualify for the ITT analysis set (e.g., fewer than 4%), approximately 135 subjects were to be randomized to each treatment group giving a total of 405 subjects. The entacapone treatment group was not included in the comparison of 40 mg/day istradefylline with placebo, but contributed to the error variance in the ANCOVA model.

No interim analysis was planned for the study.

Randomisation

Randomization (treatment) numbers were allocated sequentially by centre as subjects entered the study. The randomization scheme and associated sealed envelopes (code breaks) giving details of individual subject treatment were produced by computer software that incorporated a standard procedure for generating random numbers. Subjects were allocated to treatment in a 1:1:1 ratio in balanced blocks of three. Treatment blocks were not to be split across centres.

Blinding (masking)

Study drug was dispensed using encapsulated tablets. The istradefylline, entacapone and placebo capsules were matched for color-coding of dose time and, therefore, the subject, study staff and the Sponsor were blinded to the treatment group to which a subject was assigned. The istradefylline and placebo tablets contained in the capsules were identical in appearance, so that if the capsules were to have split, unblinding would not occur.

The blinding was not to have been broken except in emergency situations where the identification of the study treatment was specifically required for further treatment of the subject. In the event that an unblinding, the reason for breaking the code were recorded.

Statistical methods

The following analysis sets of the study populations were used: safety analysis set, ITT analysis set and PP analysis set as described for Study 6002-US-0013 and Study 6002-US-0018.

The SAP for this study was developed and finalized prior to the unblinding of treatment codes. The final SAP was issued on 04 November 2005 with an erratum issued on 22 November 2005. The database was locked on 23 November 2005 and data were unblinded for the efficacy analyses on 28 November 2005.

The primary efficacy variable was analysed using a main effects ANCOVA model with terms for Investigator and treatment as factors and Baseline as a covariate. These terms were fitted as fixed effects and remained in the model regardless of their statistical significance. The test for treatment effects was carried out from this model.

In a separate analysis, treatment by Investigator interaction was fitted into the primary main effects ANCOVA model to investigate whether treatment effect varied with Investigator. The interaction effect was tested at the two-sided alpha = 0.100. If the interaction term was significant, an assessment of whether the interaction was qualitative or quantitative was performed based on appropriate exploratory data analytic techniques and graphs.

If the interaction effect was found to be non-significant or was quantitative, the test for treatment effects from the primary main effects ANCOVA model was accepted. If the interaction term was qualitative, tests for treatment effects were assessed descriptively and the effect sizes for each Investigator were assessed for alternative explanations. Regardless of whether the interaction term was significant, the treatment effects for each Investigator were summarized with descriptive statistics for the primary efficacy variable.

In a separate analysis, treatment by Baseline interaction was fitted into the primary main effects ANCOVA model to investigate whether the assumption for parallel slopes was satisfied. The interaction term was tested at the two-sided alpha = 0.050 level. If the interaction term was significant, an assessment of whether the interaction was qualitative or quantitative was performed based on graphs. If the interaction was not significant or if the interaction was quantitative, the ANCOVA model was accepted. If the interaction term was qualitative, a main effects ANOVA model with terms for Investigator and treatment was used for the primary analysis.

The underlying assumptions of the main effects ANCOVA model were examined for the primary efficacy variable. The Shapiro-Wilk statistic was used to test for normality at the two-sided alpha = 0.05 level. Levene's test was used to test for homogeneity of variances between the treatment groups at the two-sided alpha = 0.05 level. If substantial deviations from the assumptions were found, the normalized (using Tukey's) rank transformation approach to the same ANCOVA model (without Tukey's rank transformation for the Baseline covariate) was performed to corroborate the conclusions from the primary parametric analysis.

For purposes of the analysis method described below, a pooling strategy for Investigators was employed. Data from Investigators with fewer than 6 randomized subjects were pooled based on geographic region and the pooling strategy was determined prior to the database lock and unblinding of the treatment codes.

A PP analysis was performed as a secondary analysis for the primary efficacy variable. Any substantial differences between conclusions based on the ITT analysis set compared with the PP analysis set were investigated. The PP analysis was also summarized for change from baseline in the total hours of awake time per day spent in the OFF state.

Interpretation of p-values for the primary efficacy variable at assessment times other than Endpoint and all supportive and secondary efficacy variables at all assessment times were descriptive. In addition to the p-values, 95% CIs for the differences in change from Baseline for continuous variables and in percentage of subjects for categorical variables were provided for the istradefylline and placebo groups. Additionally, for change from baseline summaries, the 95% CIs for the within treatment group change were provided.

A sensitivity analysis using an ANCOVA model for repeated measures with terms for Investigator, treatment, time, and treatment-by-time interaction as factors, and Baseline as a covariate was also conducted for the primary efficacy variable and for change from Baseline in the total hours of awake time per day spent in the OFF state to further evaluate the robustness of the results. The time factor in this model included Weeks 2, 4, 6, 8, 12, and 16.

The unstructured covariance was used for the repeated measures on each subject. This repeated measures model was an alternative approach to the LOCF method for dealing with missing data.

While the analysis of efficacy variables at Endpoint was based on the LOCF approach, the analyses at Weeks 2, 4, 6, 8, 12, and 16 were based on the observed data at the corresponding week.

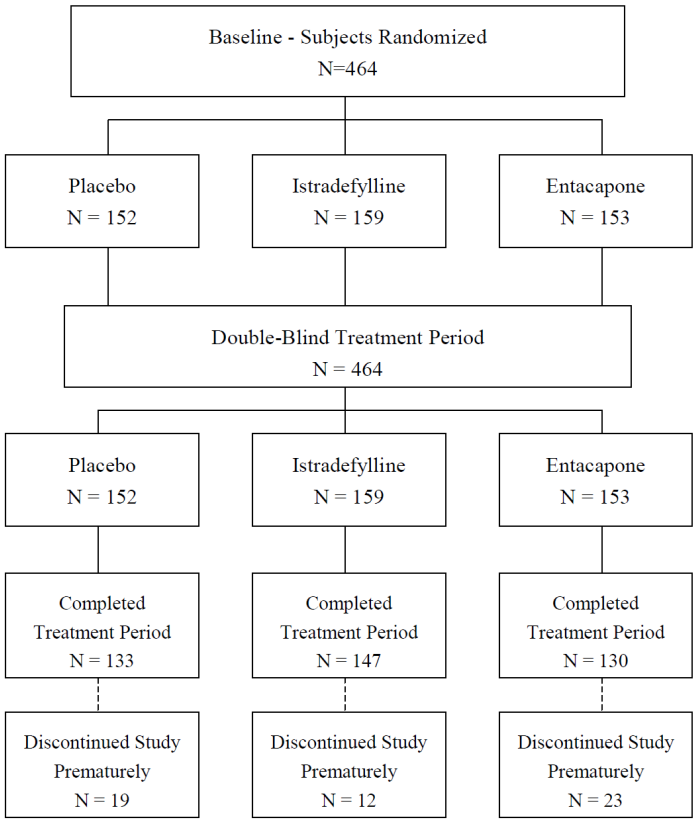
All continuous supportive and secondary efficacy variables were analysed using the main effects ANCOVA model. Tests for interactions were only summarized on these variables at Endpoint, and no further analyses were conducted, no matter whether any interactions were significant for these variables. Tests for the assumptions of the ANCOVA model were performed on these variables at Endpoint, and no further analyses were conducted, regardless of whether substantial deviations from the assumptions were found for these variables.

The CGI-S, change from Baseline for CGI-S (categorical summaries), and PGI-I variables were analysed using a Cochran-Mantel-Haenszel test using modified ridit scores and stratifying by Investigator.

Results

Participant flow

Figure 32: Disposition of subjects



Recruitment

Study locations: A total of 56 centers: Argentina (6); Austria (1); Chile (3); Estonia (3); France (1); India (7); Italy (4); Latvia (2); Lithuania (2); Russia (7); Republic of South Africa (9); Spain (4); United Kingdom (3); Ukraine (4).

Study period: first dose of study drug was on 24 November 2004 and last dose was 03 October 2005

Conduct of the study

Protocol amendment

There were 3 protocol amendments, the first was approved prior to study enrolment. Protocol amendment no.2 included allowing for modification of timing of dose of study drug and further emphasised need to take study drug with food, and otherwise changes were not major. The 3rd amendment only applied to centres in Austria, France and India as all other centres had completed the study by that time. This 3rd Amendment included allowing antipsychotic treatment for subjects experiencing levodopa- or Parkinson's disease-induced hallucinations with insight retained, and removed requirement for weight change of 3.2kg or more to be reported as AEs and otherwise changes were not major.

Protocol Deviations

In Russia, the randomisation treatment blocks were split across centres in error. According to the applicant, this was not believed to have had any effect on the results of the study.

It is unclear whether there were any major protocol deviations. The applicant has stated that none of the deviations related to study entry criteria were considered to have had a meaningful impact on study results.

Baseline data

Treatment groups were reasonably similar in terms of demographic features and baseline PD characteristics. Mean time since diagnosis was slightly longer for placebo than either drug treatment arm) as was mean duration of levodopa treatment. Mean time since onset of motor complications was similar across the groups. The mean age from 60.8 years to 62.4 years was slightly younger than that of the Study 6002-14 which also recruited a European population. Over 60% of participants were male which was similar to the other pivotal studies apart from those performed in Japan where a majority were female. Mean time since onset of motor complications was just over 3 years in all treatment groups. Mean total hours awake spent in OFF state per day varied from 6.17 hours per day in the istradefylline group, 6.47 hours in the entacapone group and 6.6 hours in the placebo group. UPDRS 1, 11, 111, IV subscale scores were similar across the groups as were the Y&H scores, PDQ 39 and PDQ 8 scores

Table 65: Baseline Demographic and PD characteristics

	Placebo N = 153 (%)	Istradefylline 40mg N = 159(%)	Entacapone N = 153(%)
Age years Mean (SD) Median (Min, Max)	 62.4 (8.6) 63 (39,80)	 60.8 (9.29) 61 (35, 81)	 61.3 (10.24) 62 (34, 87)
Male Female	92 (60.5) 60 (39.5)	99 (62.3) 60 (37.7)	93 (60.8) 60 (39.2)
Diagnosis duration years Mean (SD) Median (Min, Max)	N = 152 8.86 (4.716) 8.15 (0.6, 22.8)	N = 158 8.08 (3.903) 8 (1.4, 22.8)	N = 153 7.86 (4.503) 7.8 (0.2, 29.4)
Duration levodopa Rx yr Mean (SD) Median (Min, Max)	N = 151 7.52 (4.315) 6.9 (1.1, 21)	N = 157 1.87 (3.864) 6.7 (1, 22.8)	N = 153 7.12 (4.365) 6.4 (0.9, 28.4)
Duration motor complications years Mean (SD) Median (Min, Max)	N = 151 3.03 (2.808) 1.9 (0.2, 15.8)	N = 158 3.01 (2.376) 2.5 (0.2, 15.9)	N = 153 3.17 (3.170) 2.6 (0.1, 18.9)
Total hours awake spent in OFF state Mean (SD) Median (Min, Max)	N = 152 6.6 (2.125) 6.5 (2.3, 14)	N = 159 6.17 (2.046) 5.8 (2, 12)	N = 150 6.47 (2.346) 6 (2, 12.8)
% awake time spent in OFF state Mean (SD) Median (Min, Max)	 41.45% (12.226) 41.6% (14.8, 82.4)	 38.63 (11.592) 38.4% (13.8, 68.6)	 40.02 (13.271) 39% (13.1, 73.9)
Total hours awake spent ON without troublesome dyskinesia Mean (SD) Median (Min, Max)	N= 152 8.57 (2.297) 8.8 (0, 14)	N =159 9.11 (2.233) 9.3 (0.5, 13.3)	N = 150 9 (2.467) 8.9 (0.5, 14.8)
% awake ON state without troublesome dyskinesia Mean (SD) Median (Min, Max)	N = 152 54.07% (13.513) 55.55% (0, 81.5)	N =159 57.21% (12.874) 58% (5, 83.7)	N = 150 56.19% (14.477) 56.7% (0, 86.9%)

H&Y in OFF state 2 2.5 3 4	N = 152 24 (15.8%) 46 (30.3%) 65 (42.8%) 17 (11.2%)	N =159 27 (17%) 52 (23.7%) 65 (40.9%) 15 (9.4%)	N = 153 24 (15.7%) 46 (30.1%) 65 (42.5%) 18 (11.8%)
Concomitant medications Note: All patients were taking carbidopa/levodopa or benserazide levodopa combination			
Levodopa Daily dose mg Mean (SD) Median (Min, Max)	N = 152 649 (344.23) 600 (188, 2000)	N =159 608 (303.24) 509.5 (105, 1638)	N =153 635.4 (308.92) 600 (188, 1716)
Amantadine	39 (25.7%)	49 (30.8%)	49 (32%)
Ropinirole (DA)	30 (19.7%)	29 (18.2%)	22 (14.4%)
Pramipexole (DA)	22 (14.5%)	27 (17%)	24 (15.7%)
Selegiline (MAO-B inhibitor)	16 (10.5%)	11 (6.9%)	11 (7.2%)
Bromocriptine (DA ergot)	7 (4.6%)	6 (3.8%)	16 (10.5%)
Pergolide (DA ergot)	8 (5.3%)	5 (3.1%)	5 (3.3%)

Numbers analysed

Of the 464, one hundred and fifty-two were randomised to receive placebo, 159 to receive istradefylline 40 mg/day, and 153 to receive entacapone. A total of 410 patients completed the study: 147 (92.5%) in the istradefylline group, 130 (85%) in the entacapone group, and 133 (87.5%) in the placebo group. AEs were the most common reason for premature discontinuation from the study with 7 (4.4%) in the istradefylline group, 10 (6.5%) in the entacapone group, and 10 (6.6%) in the placebo group.

Table 66: Study Participant disposition

Patient disposition			
	Placebo N (%)	Istradefylline 40mg N (%)	Entacapone N (%)
Randomised Safety analysis set[b] ITT[c] PP[d]	152 152 151 (9.3) 125 (82.2)	159 159 158 (99.4) 141 (88.7)	153 153 146 (95.4) 124 (81)
Completed 16 weeks	133 (87.5)	147 (92.5)	130 (85)

b All randomized subjects who took at least 1 dose of double-blind study drug.
c All randomized subjects who took at least 1 dose of double-blind study drug and had a Baseline and at least 1 post-Baseline 24-hour ON/OFF Parkinson's disease patient diary.
d Subjects who are in the ITT analysis set and who completed 16 weeks of treatment with no major protocol violations.

Outcomes/endpoints

Primary efficacy endpoint

For the primary efficacy endpoint the percentage reduction in OFF time at endpoint from baseline was similar for placebo and istradefylline (ITT) (-4.76% v -4.31% respectively). The change for entacapone was numerically greater (-7.42%) (

Table 67). The outcomes by time point are also shown in the accompanying Figure which show that change from baseline at endpoint was broadly similar for the placebo and istradefylline at the various timepoints with a greater separation between placebo/istradefylline and entacapone.

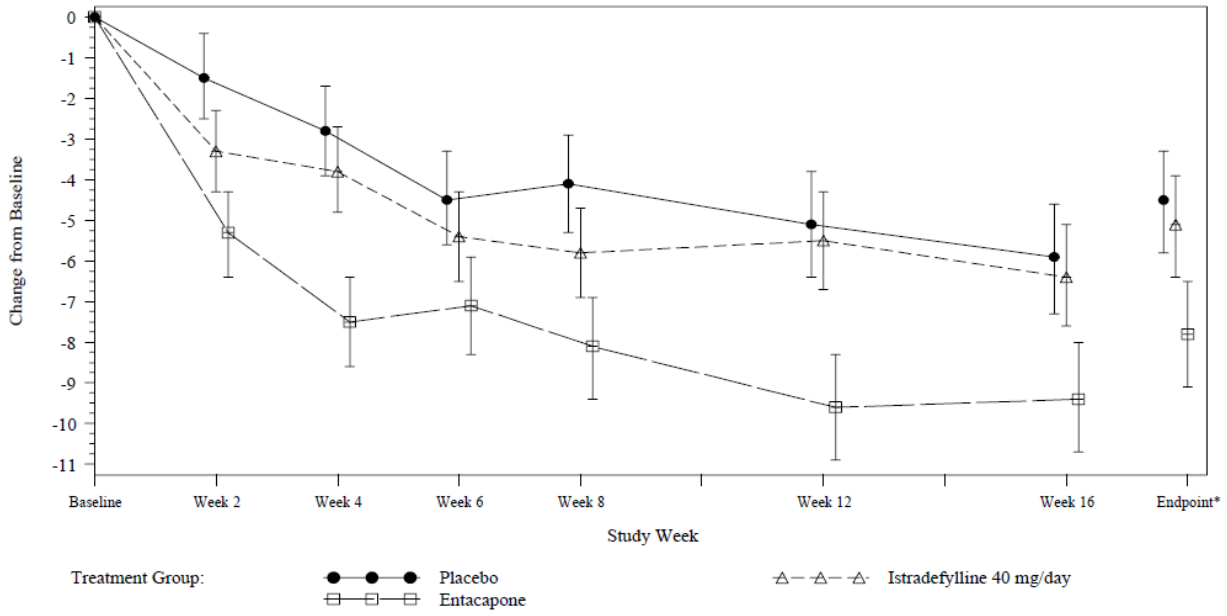
A worst case scenario analysis was also conducted which showed broadly similar results.

Table 67: Percentage of awake time per day spent in an OFF state (observed case analysis)

	Placebo N = 151	Istradefylline 40mg N = 158	Entacapone N = 146
Baseline [b] Mean (SD) Median (min, max)	 41.49 (12.257) 41.7 (14.8, 82.4)	 38.61 (11.625) 38.35 (13.8, 68.6)	 40.05 (13.423) 39 (13.1, 73.9)
Endpoint Mean (SD) Median (min, max)	 36.73 (17.311) 37.8 (0, 81.1)	 34.3 (16.095) 35.05 (0, 73.9)	 32.63 (17.741) 31.65 (0, 75.4)
Change at endpoint from baseline Mean (SD) Median (min, max) LSM (95% CI) LSM diff v placebo (95% CI) P value (versus placebo)[c]	N = 151 -4.76 (15.858) -1.2 (-82.4, 34.4) -4.53 (-7.02, -2.04)	N = 158 -4.31 (15.544) -3.75 (-62.5, 33.9) -5.14 (-7.58, -2.69) -0.61 (-4.05, 2.83) 0.729	N =146 -7.42 (16.358) -4.95 (-50.7, 33.4) -7.82 (-10.34, -5.3) -3.29 (-6.77,0.19) 0.064

a Based on valid 24-hour ON/OFF patient diary from observed-case analysis.
b Baseline values were collected from diaries dispensed at Day -1. Subjects with more than 4 invalid entries in any 1 of their 2 daily diaries had to repeat the Baseline (Day -1) visit 7 days later. Subjects failing the validity criterion for a second time were precluded from participating in the study.
c The pairwise p-value is based on LSM from main effects ANCOVA with terms for Baseline, Investigator, and treatment.

Figure 33: Primary Efficacy: Change from Baseline by Study Visit and at Endpoint in the percentage of Awake Time per Day Spent in the OFF State (observed case analysis)



Secondary Efficacy variables

This study failed to demonstrate any efficacy for istradefylline 40mg over placebo in the primary endpoint. Therefore, results for secondary efficacy endpoints are not further considered (percentage of awake time / day spent ON without troublesome dyskinesia is provided in the table below for completeness as a relevant secondary efficacy endpoint).

Subgroup analyses

A number of sub-group analyses were performed. The study failed to demonstrate efficacy in the primary endpoint. Subgroup results are not displayed.

Summary of main efficacy results

This summary should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 68: Summary of efficacy for trial 6002-EU-007

Title: A 16-week, Double-Blind, Placebo-Controlled, Randomised, Parallel-Group, Multicentre, International Study to Evaluate the Efficacy and Safety of 40 mg/day KW-6002 (istradefylline) and that of Entacapone versus Placebo as Treatment for Parkinson's Disease in Patients with Motor Complications on Levodopa Therapy					
Study identifier	6002-EU-007				
Design	Phase 3, randomised, double-blind, placebo-controlled, fixed-dose, parallel-group, multicenter trial				
	Duration of main phase: Duration of Run-in phase:		16 weeks Not applicable		
	Duration of Extension phase:		Not applicable		
Hypothesis	Superiority				
Treatments groups	Istradefylline 40 mg		Istradefylline 40 mg once daily, 16 weeks, n=159 subjects		
	Entacapone 200 mg/levodopa dose		Entacapone 200 mg with each dose of levodopa, 16 weeks, n=153 subjects		
	Placebo		Placebo once daily, 16 weeks, n=152 subjects		
Endpoints and definitions	Primary endpoint	OFF time per day	Change from baseline in the percentage of awake time/day spent in the OFF state		
	Key secondary endpoint	ON time per day without troublesome dyskinesia	Change from baseline in percentage of awake time/day spent ON without troublesome dyskinesia		
Database lock	23 November 2005				
Results and Analysis					
Analysis description	**Primary Analysis**				
Analysis population and time point description	Intent to treat (ITT), week 16/Endpoint LOCF, Observed case, ANCOVA				
Descriptive statistics and estimate variability	Treatment group		Placebo	Istradefylline 40 mg	Entacapone 200mg/LD dose
	Number of subjects analysed at Week 16/ Endpoint		151	158	146
	Change from baseline in the percentage of awake time/day spent in the OFF state; Mean (SD)		-4.76 (15.858)	-4.31 (15.544)	-7.42 (16.358)
	Range (min to max)		-82.4 to 34.4	-62.5 to 33.9	-50.7 to 33.4

	Change from baseline in percentage of awake time/day spent in the ON state without troublesome dyskinesia; Mean (SD)	5.49 (17.677)	3.39 (15.626)	5.90 (17.937)
	Range (min to max)	-39.0 to 82.4	-50.3 to 46.1	-50.4 to 48.7
Effect estimate per comparison	Primary endpoint (% of awake time/day spent in the OFF state); difference from placebo in the LSM change from baseline	Comparison groups	Istradefylline vs Placebo	Entacapone vs Placebo
		LSM change vs. placebo	-0.61	-3.29
		95% CI	-4.05, 2.83	-6.77, 0.19
		P-value	0.729	0.064
	Key secondary endpoint (% of awake time/day spent ON without troublesome dyskinesia); difference from placebo in the LSM change from baseline	Comparison groups	Istradefylline vs Placebo	Entacapone vs Placebo
		LSM change vs. placebo	-0.88	1.31
		95% CI	-4.62, 2.87	-2.49, 5.10

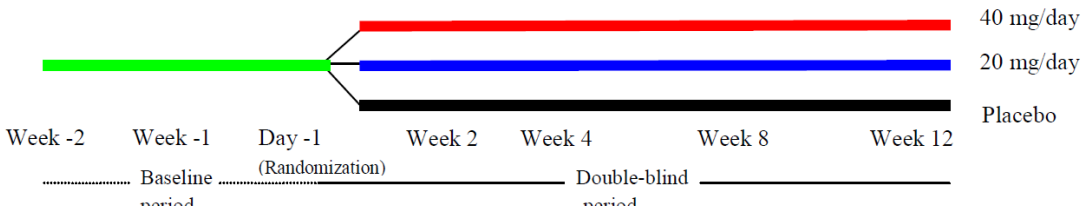
Study 6002-0608

"A phase 2 randomised placebo controlled study conducted in Japan with 12 weeks treatment duration, entitled Placebo-controlled, double-blind, parallel group, fixed dose study of KW-6002 (Istradefylline) in the treatment of Parkinson's Disease"

Methods

Study design

Figure 34: study design of Study 6002-0608



Study Participants

Inclusion criteria

- Aged 20 years or more diagnosed with PD based on UKPDS Brain bank criteria for PD.
- PD in Stages 2-4 modified H&Y Scale while in an OFF state on the day of randomisation (D -1).
- Had responded to levodopa/DCI (carbidopa or benserazide) and had been treated for at least 1 year prior to randomisation, were taking at least 3 doses of levodopa/DCI per day with a daily dose of at least 300mg within 4 weeks prior to randomisation and with predictable end of dose wearing off.
- Successful completion of diary training.

- Had an average of at least 2 hour of OFF time per day on at least 4 valid diaries completed during 7 days prior to randomisation after successfully completing diary training.
- No new anti PD drugs and a stable regiment of anti-PD drugs for at least 4 weeks prior to randomisation

Main Exclusion criteria

- Treatment with CYP3A4 substrates or inhibitors or zonisamide within 2 weeks prior to randomisation.
- Treated with MAO inhibitors except selegiline within 3 months prior to randomisation
- Treated within 3 months prior to randomisation with typical or atypical antipsychotics (6 months for depot formulations), centrally acting dopamine antagonists, reserpine and papaverine
- Neurosurgical operation for PD (stereotactic surgery, or deep brain stimulation) at any time; or transcranial magnetic stimulation within 6 months prior to randomisation
- Prior exposure to KW-6002.
- History of dementia or a score of ≤ 25 on the MMSE conducted during the baseline period.
- History of psychotic illness, apart from psychiatric symptoms related to PD

Treatments

There were 3 treatment groups:

- 20mg KW-6002 (2 x 10mg tablets in am once daily for 12 weeks)
- 40 mg KW-6002 (2 x 20mg tablets in am once daily for 12 weeks)
- Placebo (2 x placebo tablets in am once daily for 12 weeks)

Changes in existing stable anti PD drug regimens were not allowed in the course of the study but could be modified (decreased) in repose to Treatment emergent adverse events (TEAE). Prophylactic use of domperidone was not allowed unless for the treatment of a TEAE.

Objectives

The objectives of the study were to:

- evaluate the efficacy of KW-6002 at a PO dose of 20mg or 40mg daily for 12 weeks in PD patients with motor complication on levodopa therapy based on the change in total hours of awake time spent in the OFF state per day compared to placebo
- determine the recommended clinical dose
- assess safety

Outcomes/endpoints

Primary endpoints

Change in the total hours of awake time per day spent in the OFF state at the last available post baseline visit. The total hours of awake time per day spent in the OFF state was defined as the number of 30-minutes periods classified as "OFF" in the patient's ON/OFF diary multiplied by 0.5 hours. Data on ON/OFF periods was to be completed in the daily diaries during the seven-day period before each study visit. If at least four valid diaries were not available for evaluation, the total hours of awake time per day spent in the OFF state at that assessment time were to be set to missing.

Secondary endpoints

The following endpoints were to be determined at each post baseline assessment visit with primacy given to the last post baseline visit value.

- Change in the percentage of awake time per day spent in the OFF state
- Change in the total hours of awake time per day spent in the ON state for each of the dyskinesia-based subcategories of ON time i.e without dyskinesia, with dyskinesia, with non-troublesome dyskinesia, with troublesome dyskinesia, without troublesome dyskinesia
- Change in the percentage of awake time per day spent in the ON state for each of the dyskinesia-based subcategories of ON time
- Change in the UPDRS Part I, Part II (ON and OFF states), Part III (ON state) and Part IV subscale scores, and Part I to III total score (ON state)
- The CGI-I.
- Change in the modified H&Y stage

Sample size

The planned sample size was a total of 360 subjects: 120 subjects each in the 20 mg/day KW-6002 group, 40 mg/day KW-6002 group, and placebo group.

Calculation of the sample size was informed by 2 RCTs one in the US (6002-US-005) and the Japanese phase 2a study (6002-0406; a DB intra-group comparison study with 20 mg/day KW-6002, 40 mg/day KW-6002, and placebo. Study 6002-US-005 was designed to determine the efficacy of KW-6002 compared with placebo. The results of an ANCOVA analysis showed that the LSM reductions in the total hours of awake time per day spent in the OFF state were 1.8 hours for 40 mg/day KW-6002 and 0.7 hours for placebo, with a difference of 1.1 hours and a standard deviation of 2.5 hours. In Study 6002-0406 (Japan), the results of an ANCOVA analysis showed that the LSM reductions from Baseline in the total hours of awake time per day spent in the OFF state for subjects treated with 40 mg/day KW-6002 were 1.5 hours when analysed based on the FAS; the LSM reductions in the total hours of awake time per day spent in the OFF state for placebo-treated subjects were 1.1 hours based on the FAS. The SD of the error was estimated to be 2.7 hours based on the FAS. In Study 6002-0406, the total hours of awake time per day spent in the OFF state for 40 mg/day KW-6002-treated subjects were reduced to an extent similar to that in Study 6002-US-005, whereas the reductions in the total hours of awake time per day spent in the OFF state for placebo-treated subjects were 1.5 to 2 times greater than those in Study 6002-US-005.

Randomisation

Subjects were to be randomised to one of the treatment groups using the permuted block method in which subjects were divided into blocks by investigative site. The treatment assignment manager was to randomly assign the three groups (20 mg/day KW-6002, 40 mg/day KW-6002, and placebo) in each block. Study drug for each subject was to be sealed. In order of randomization, the investigator/subinvestigator was to allocate to the subject the lowest Subject No. in the block upon randomization, and dispense the assigned study drug.

Blinding (masking)

This was a double blind trial. Before the start of the treatment assignment process, the treatment assignment manager verified that the randomly sampled investigational drug and comparator were identical in appearance, shape, odour, package, label and other characteristics (on 23 February 2007).

In addition, before the post-study opening of the allocation table, the treatment assignment manager reviewed the samples retained for the verification of study drug tablets being identical in appearance and confirmed that it was impossible to distinguish between the investigational drug and comparator based on appearance, shape, odour, package, label and other characteristics (on 25 September 2008).

Based on these results, it was determined that study drug tablets used in the 20 mg/day KW-6002 group, 40 mg/day KW-6002 group and placebo group had been identical in appearance and that both study personnel and subjects had been blinded to treatment assignment throughout the study.

Statistical methods

The primary and secondary efficacy endpoints were evaluated in the full analysis set population (FAS) i.e. all those who were randomised who received at least one dose of study drug and who submitted at least 4 valid diaries at any post baseline assessment time

An ANCOVA model was to be used for the main analysis of the total hours of awake time per day spent in the OFF state. An ANCOVA model with terms for treatment as a factor and baseline value and study centre as covariates was to be fitted to the change from Baseline in the total hours of awake time per day spent in the OFF state at each post-Baseline assessment time or Endpoint. In the ANCOVA model, the LSM and its 95% CIs were to be calculated for each treatment group to estimate the change in the total hours of awake time per day spent in the OFF state. In addition, the LSM difference from placebo and its 95% CIs were to be calculated for 20 mg/day and 40 mg/day KW-6002.

To determine the efficacy of KW-6002 relative to placebo at Endpoint, 20 mg/day and 40 mg/day KW-6002 were to be compared with placebo using Williams' test based on the assumption that there would be a dose-response relationship. This test was to be conducted at the one-sided significance level of 2.5%.

The study centres to be used in the ANCOVA model were to be defined by investigator. If the number of subjects was so small that some centres needed to be combined, they were to be determined through the blinded review to be conducted before database lock.

As with the analysis of the total hours of awake time per day spent in the OFF state, an ANCOVA model with terms for the change from Baseline as a response variable and Baseline value and study centre as covariates was to be fitted to the percentage of awake time per day spent in the OFF state, total hours and percentage of awake time per day spent in the ON state for each of the subcategories of ON time, and UPDRS scores. The LSM and its 95% CIs were to be calculated for each treatment group. In addition, the LSM and its 95% CIs were to be calculated for 20 mg/day and 40 mg/day KW-6002.

For the CGI-I, the percentage of subjects classified as each of the improvement categories at each post-Baseline assessment time or Endpoint was to be calculated by treatment group. In addition, the percentages of subjects in the "Much improved" or better category ("Very much improved" + "Much improved") and in the "Minimally improved" or better categories ("Much improved" or better + "Minimally improved") were to be calculated by treatment group.

For the modified H&Y staging, the change in score from Baseline to Week 12/ET in each treatment group was to be summarized using shift tables.

If data at Week 12 (or ET) were missing in the analyses of the primary and secondary efficacy variables, the LOCF approach was used, i.e., the last available post-Baseline value was used as Endpoint.

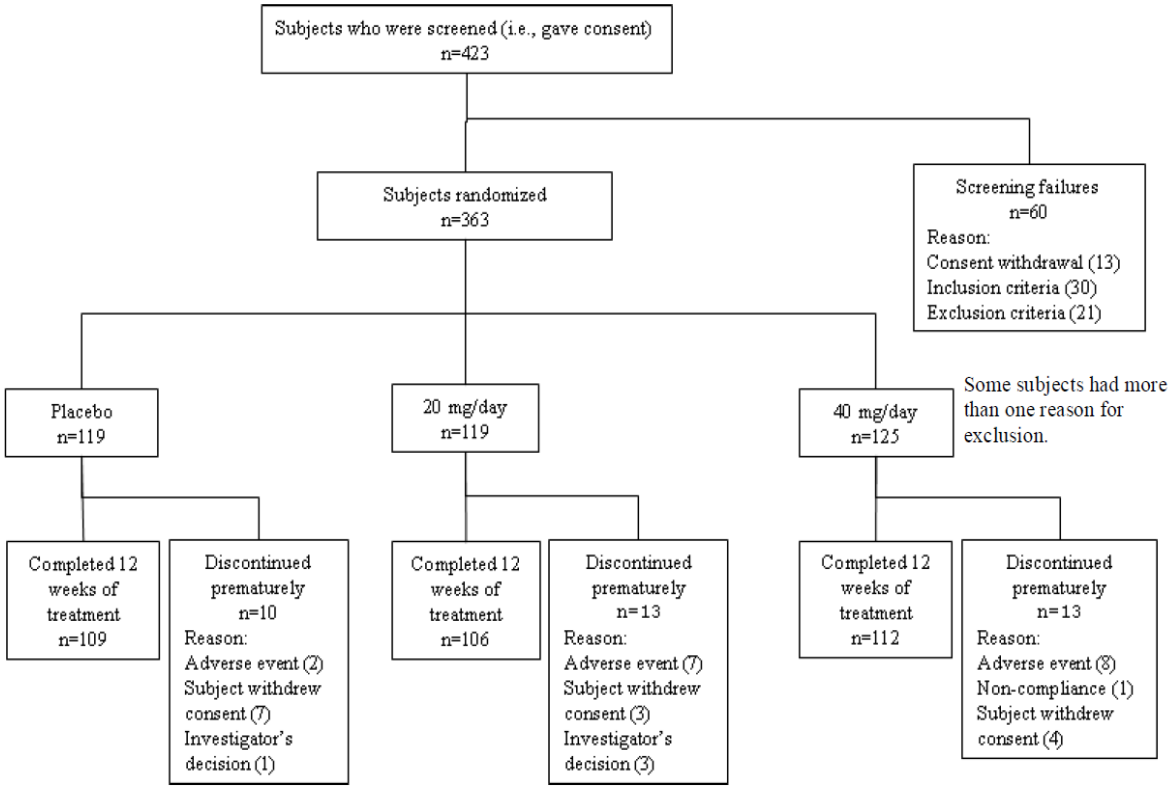
For patient's ON/OFF diaries, an observed case analysis was primary, i.e., 30-minutes periods with invalid entries were treated as missing and not included in the analysis. The stability of the observed case analysis was assessed by a worst-case analysis, in which 30-minute periods with missing entries were imputed with OFF, and those periods with multiple entries were imputed with the worst-case value (the ordering for selecting the worst-case was based on the following: "Asleep" < "ON state without dyskinesia," < "ON state with non-troublesome dyskinesia," < "ON state with troublesome dyskinesia" < "OFF").

To determine the effect of subject withdrawal in the early stage of treatment, the change from Baseline in the total hours of awake time per day spent in the OFF state (observed case analysis), which was the primary variable, was subjected to the following analysis without imputation: all available data between Week 2 and Week 12 were analysed by a repeated measures ANCOVA model with terms for the main effect of time and treatment-by-time interaction added to the model. The model was fitted based on the assumption that the variance-covariance structure of errors was Compound Symmetry.

Results

Participant flow

Figure 35: Disposition of subjects



Four hundred and twenty-three patients were screened of whom 363 were recruited to the study. Completion rates at 12 weeks were high and similar across treatment groups (at least 89%).

Recruitment

Study location: Japan

Study period: first patient entered the study on 30 March 2007 and the last patient exited the study on 5 August 2008

Conduct of the study

Protocol amendment

There appear to have been no major changes to the protocol during the duration of the study. The study protocol was revised four times (ver.2. 02.04.2007; ver.3 01.11.2007; ver.4 10.03.2008; ver.5 16.04.2008) mainly to change the date of the last randomization and extension of the study duration.

Protocol Deviation

Ten protocol deviation were reported by the applicant as significant related with the use of restricted / prohibited concomitant medications (n=7) or non-compliance (n=3).

Baseline data

The mean age was 65.0 years (range 38 to 84 years) for the placebo group, 65.1 years (49 to 81 years) for the 20 mg/day KW-6002 group, and 63.7 years (37 to 80 years) for the 40 mg/day KW-6002 group. Male subjects accounted for 38.1% (placebo), 43.5% (Istradefylline 20mg), and 44.4% (Istradefylline 40mg) of the population. The mean time since diagnosis of PD was 8.338 years, 8.037 years, and 8.089 years, respectively. The mean time since onset of motor complications was 3.506 years, 3.167 years, and 3.126 years, respectively.

Based on patient's ON/OFF diaries, the mean total hours of awake time per day spent in the OFF state were 6.43 hours, 6.79 hours, and 6.55 hours, respectively, the mean percentages of awake time per day spent in the OFF state were 39.50%, 41.07%, and 39.79%, respectively, and the mean total asleep hours per day were 7.80 hours, 7.57 hours, and 7.49 hours, respectively.

Table 69: demographic and baseline characteristics (full analysis set)

	Placebo N = 118	Istradefylline 20mg/day N =115	Istradefylline 40mg/day N = 124
Age Mean (SD) Median (min, max) ≥ 65	 65 (7.6) 65 (38, 84) 61 (51.7%)	 65.1 (7.2) 66 (49, 81) 66 (57.4%)	 63.7 (8.6) 64 (37, 80) 60 (48.4%)
Gender F/M	61.9%/38.1%	56.5%/43.5%	55.6%/44.4%
Duration of diagnosis (years) Mean (SD) Median (min, max) ≥ 10 years	 8.338 (4.826) 7.77 (1.06, 30.65) 37 (31.4%	 8.037 (4.076) 7.66 (1.41, 18.93) 35 (30.4%)	 8.089 (4.048) 7.4 (0.39, 21.17) 34 (27.4%)
Time since onset motor complics (years) Mean (SD) Median (min, max)	 3.506 (3.015) 2.72 (0.1, 13.19) 36 930.5%)	 3.167 (2.499) 2.69 (0.11, 13.2) 40 (34.8%)	 3.126 (2.884) 2.325 (0.05, 19.15) 38 (30.6%)

≥ 4 years			
Hours awake spent in OFF state Mean (SD) Median (min, max) ≥ 6 hours	 6.43 (2.71) 6.3 (2, 14.8) 64 (54.2%)	 6.79 (2.86) 6.6 (2.2, 17) 68 (59.1%)	 6.55 (2.48) 6.35 (2.1, 14) 73 (58.9%)
On without dyskinesia hours Mean (SD) Median (min, max)	 7.58 (3.47) 8.05 (0, 15.3)	 7.79 (3.48) 8.3 (0, 14.9)	 8.01 (3.51) 8.35 (0, 15.5)

Over 90% of patients were taking concomitant PD medication. Eighty-nine percent in the placebo arm, 95.7% in the Istradefylline 20mg am and 91.9% in the Istradefylline 40mg arm were taking a dopamine agonist of which the commonest was pramipexole which was used by at least 50% of all the treatment groups (See Table 70). A similar proportion used selegiline. There was higher use of entacapone in the Istradefylline 20mg group (19.1%) compared to the placebo (12.7%) and istradefylline 40mg (12.9%) groups.

Table 70: Concomitant medications (full analysis set)

Variable			Treatment	Placebo	20mg/day	40mg/day	p-value[a]
			n	118	115	124	
Concomitant medication[d]			Yes	107(90.7%)	110(95.7%)	112(90.3%)	b)0.237
			No	11(9.3%)	5(4.3%)	12(9.7%)	
Daily Dosage of Prior Levodopa			Mean+/-Std Dev	426.3 ±143.0	407.0 ±113.1	415.3 ±159.2	c)0.726
			Median	400.0	400.0	400.0	
			Minimum, Maximum	300, 900	300, 700	300, 1500	
			-300	41(34.7%)	40(34.8%)	48(38.7%)	b)0.710
			300<-600	66(55.9%)	69(60.0%)	68(54.8%)	
			>600-	11(9.3%)	6(5.2%)	8(6.5%)	
Concomitant Antiparkinsonian Drugs	Levodopa Mono		Yes	0(0.0%)	0(0.0%)	0(0.0%)	b)-
			No	118(100%)	115(100%)	124(100%)	
	DOPAMINE AGONISTS		Yes	105(89.0%)	110(95.7%)	114(91.9%)	b)0.166
			No	13(11.0%)	5(4.3%)	10(8.1%)	
	DOPAMINE AGONISTS	BROMOCRIPTINE	Yes	5(4.2%)	0(0.0%)	1(0.8%)	b)0.027+
			No	113(95.8%)	115(100%)	123(99.2%)	
		PERGOLIDE	Yes	22(18.6%)	29(25.2%)	31(25.0%)	b)0.394
			No	96(81.4%)	86(74.8%)	93(75.0%)	
		CABERGOLINE	Yes	32(27.1%)	24(20.9%)	32(25.8%)	b)0.506
			No	86(72.9%)	91(79.1%)	92(74.2%)	
		TALIPEXOLE	Yes	1(0.8%)	7(6.1%)	4(3.2%)	b)0.085+
			No	117(99.2%)	108(93.9%)	120(96.8%)	
		PRAMIPEXOLE	Yes	66(55.9%)	68(59.1%)	62(50.0%)	b)0.353
			No	52(44.1%)	47(40.9%)	62(50.0%)	
		ROPINIROLE	Yes	6(5.1%)	5(4.3%)	7(5.6%)	b)0.900
			No	112(94.9%)	110(95.7%)	117(94.4%)	
	ANTICHOLINERGIC AGENTS		Yes	23(19.5%)	18(15.7%)	23(18.5%)	b)0.729
			No	95(80.5%)	97(84.3%)	101(81.5%)	
	SELEGILINE		Yes	62(52.5%)	57(49.6%)	67(54.0%)	b)0.782
			No	56(47.5%)	58(50.4%)	57(46.0%)	
	ENTACAPONE		Yes	15(12.7%)	22(19.1%)	16(12.9%)	b)0.292
			No	103(87.3%)	93(80.9%)	108(87.1%)	
	DROXIDOPA		Yes	12(10.2%)	3(2.6%)	14(11.3%)	b)0.030+
			No	106(89.8%)	112(97.4%)	110(88.7%)	
	AMANTADINE		Yes	45(38.1%)	43(37.4%)	39(31.5%)	b)0.491
			No	73(61.9%)	72(62.6%)	85(68.5%)	
	Others		Yes	0(0.0%)	0(0.0%)	0(0.0%)	b)-
			No	118(100%)	115(100%)	124(100%)	
Combinations of Prior Antiparkinsonian Drugs [e]			Levodopa only	2(1.7%)	1(0.9%)	2(1.6%)	b)0.982
			Levo+DA	28(23.7%)	27(23.5%)	32(25.8%)	
			Levo+DA+SEL/ENT	38(32.2%)	41(35.7%)	46(37.1%)	
			Levo+DA+SEL/ENT+AMA	25(21.2%)	25(21.7%)	24(19.4%)	
			Others	25(21.2%)	21(18.3%)	20(16.1%)	

a +p<0.15; b chi-square; test c Kruskal-Wallis test; d Concomitant medications except antiparkinsonian drugs that were taken within 4 weeks prior to randomization; e DA=Dopamine agonists, SEL=Selegiline; ENT=Entacapone, AMA=Amantadine

Numbers analysed

Overall the FAS represented at least 98% of those randomised. Completion rates at 12 weeks were high from 89.1% to 91.6%.

For the variables derived from patient's ON/OFF diaries, the following two approaches were used to handle 30-minute periods with invalid entries (more than one check mark or absence of an entry). The observed case analysis was primary analysis.

- Observed case analysis: Invalid entries were set to missing and not included in the analysis.

- Worst-case analysis: 30-minute periods with missing entries were imputed with OFF, and those periods with multiple entries were imputed with the worst-case value (the ordering for selecting the worst-case was based on the following: Asleep < ON state without dyskinesia < ON state with non-troublesome dyskinesia < ON state with troublesome dyskinesia < OFF).

Table 71: Disposition of patients

	Placebo	20mg/day	40mg/day
Randomised	119	119	125
Treated	119	118 (99.2%)	125
Completed			
Week 2	117 (98.3%)	116 (97.5%)	124 (99.2%)
Week 4	115 (96.6%)	112 (94.1%)	122 (97.6%)
Week 8	112 (94.1%)	107 (89.9%)	118 (94.4%)
Week 12	109 (91.6%)	106 (89.1%)	112 (89.6%)
Full analysis set *	118 (99.2%)	115 (96.6%)	124 (99.2%)
Per protocol	106	105	109

FAS = Subjects who were randomized; who received at least one dose of study drug; and who submitted at least four valid diaries subject to evaluation at any of the post-Baseline assessment times.

Outcomes and estimation

Primary endpoint

The primary variable in this study was the difference between the total hours of awake time per day spent in the OFF state at each post-Baseline assessment time and those at Baseline. The primary assessment time was the time of the last available post-baseline value (Endpoint) for each subject.

The LSM changes from Baseline at Endpoint in the total hours of awake time per day spent in the OFF state estimated based on the ANCOVA model were -0.66 hours (95% C.I -1.08 to -0.25 hours) for the placebo group, -1.31 hours (-1.73 to -0.89 hours) for the 20 mg/day KW-6002 group, and -1.58 hours (-1.99 to -1.17 hours) for the 40 mg/day KW-6002 group.

The LSM differences from the placebo group at Endpoint in the total hours of awake time per day spent in the OFF state were -0.65 hours (95% C.I -1.23 to -0.07 hours) for the 20 mg/day KW-6002 group and -0.92 hours (-1.49 to -0.35 hours) for the 40 mg/day KW-6002 group. The results of Williams' test showed that 40 mg/day KW-6002 statistically significantly reduced the total hours of awake time per day spent in the OFF state compared with placebo ($p<0.001$). The reduction in the total hours of awake time per day spent in the OFF state in the 20 mg/day KW-6002 group was also statistically significantly greater than that in the placebo group ($p=0.013$).

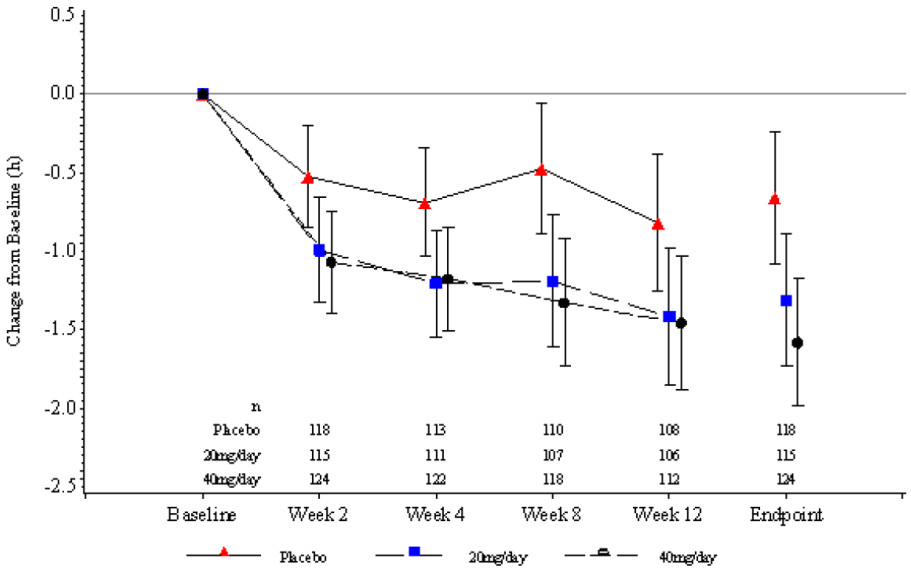
Difference in mean reduction in OFF time for Istradefylline 20mg was 39 minutes and 55 minutes for istradefylline 40mg.

Table 72: Total hours of awake time per day spent in the OFF state based on patient´s ON/OFF diary-actual and change from baseline values (observed case analysis)

			Placebo n=118		20mg/day n=115		40mg/day n=124	
			Actual	Change	Actual	Change	Actual	Change
Baseline	Statistics	n	118		115		124	
		Mean	6.43		6.79		6.55	
		Std Dev	2.71		2.86		2.48	
		Minimum	2.0		2.2		2.1	
		Median	6.30		6.60		6.35	
		Maximum	14.8		17.0		14.0	
Endpoint	Statistics	n	118	118	115	115	124	124
		Mean	5.80	-0.62	5.42	-1.36	4.99	-1.55
		Std Dev	3.27	2.17	3.37	2.01	2.68	2.66
		Minimum	0.3	-8.0	0.0	-7.1	0.0	-10.7
		Median	5.55	-0.55	4.70	-1.20	5.10	-1.30
		Maximum	15.8	7.0	14.6	3.0	14.4	5.3
	ANCOVA	LS Mean	5.92	-0.66	5.27	-1.31	5.00	-1.58
		95%C.I.		-1.08, -0.25		-1.73, -0.89		-1.99, -1.17
	ANCOVA (vs. Placebo)	LS Mean				-0.65		-0.92
		95%C.I.				-1.23, -0.07		-1.49, -0.35
		p-value				0.028		0.002
	Williams Test	Test Statistic, DF				2.23, 321		3.22, 321
		p-value				0.013 *		<.001 *

LSM and p-value are based on the main effects ANCOVA with terms for baseline, investigator and treatment
William´s test p-value * p<0.025, NS: Not significant.

Figure 36: Change from baseline (LSM and 95% CI) in the total hours of awake time per day spent in the OFF state based on patient´s ON/OFF diary-actual (observed case analysis)



Similar results were obtained in the PP and in the FAS population with a worst case analysis (Table 73).

Table 73: Total hours of awake time per day spent in the OFF state based on patient´s ON/OFF diary-actual and change from baseline values (worst case analysis)

	Placebo	20mg/day	40mg/day
Baseline Mean (SD) Median (Min, max)	N =118 6.44 (2.71) 6.3 (2, 14.8)	N =115 6.81 (2.05) 6.6 (2.2, 17)	N =124 6.56 (2.48) 6.35 (2.1, 14)
Endpoint Mean (SD) Median (Min, max)	N =118 5.81 (3.27) 5.6 (0.3, 15.8)	N = 115 5.44 (3.37) 4.7 (0, 14.6)	N = 124 5 (2.67) 5.15 (0, 14.4)
ANCOVA LSM change (95%CI)	-0.67 (-1.08, -0.25)	-1.32 (-1.74, -0.9)	-1.58 (-1.99, -1.18)
ANCOVA LSM change v placebo (95% CI)		-0.65 (-1.23, -0.07)	-0.92 (-1.49, -0.35)
Williams test P value		0.013	< 0.001

LSM and p-value are based on the main effects ANCOVA with terms for baseline, investigator and treatment
William´s test p-value * p<0.025, NS: Not significant.

Secondary endpoints

Percentage of awake time spent in the OFF state.

For this secondary endpoint the results mirrored those of the primary endpoint with a small but statistically significant reduction in the percentage time spent in the OFF state compared to placebo which was greater for istradefylline 40mg than Istradefylline 20mg (See

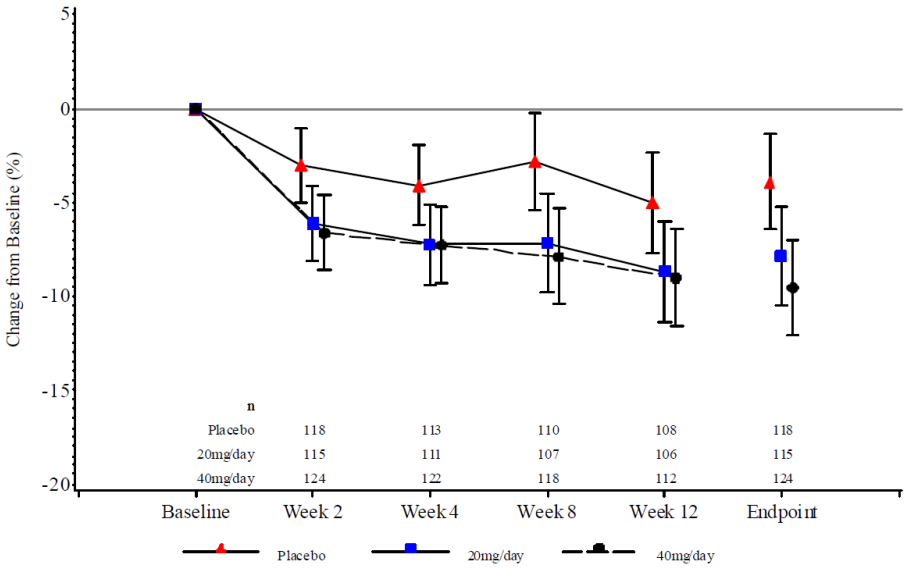
Table 74).

Table 74: Percentage of awake time per day spent in the OFF state based on on patient´s ON/OFF diary-actual and change from baseline values (observed case analysis)

		Placebo n=118		20mg/day n=115		40mg/day n=124	
		Actual	Change	Actual	Change	Actual	Change
Baseline	Statistics n	118		115		124	
	Mean	39.50		41.07		39.79	
	Std Dev	16.13		16.14		15.18	
	Minimum	11.4		12.6		13.6	
	Median	37.40		41.00		38.60	
	Maximum	90.9		92.6		90.6	
Endpoint	Statistics n	118	118	115	115	124	124
	Mean	35.74	-3.76	32.85	-8.22	30.35	-9.44
	Std Dev	19.21	13.61	19.69	12.89	16.03	16.11
	Minimum	1.7	-52.0	0.0	-54.1	0.0	-63.1
	Median	33.70	-3.70	29.60	-7.30	32.75	-6.70
	Maximum	85.7	47.4	89.5	15.5	85.7	29.3
	ANCOVA LS Mean	36.22	-3.88	32.23	-7.88	30.55	-9.55
	95%C.I.		-6.44, -1.32		-10.48, -5.28		-12.07, -7.04
	ANCOVA LS Mean				-3.99		-5.67
	(vs. 95%C.I.				-7.57, -0.41		-9.18, -2.17
	Placebo) p-value				0.029		0.002
	Williams Test				2.22, 321		3.21, 321
	Statistic, DF						
	p-value				0.014 *		<.001 *

LSM and p-value are based on the main effects ANCOVA with terms for baseline, investigator and treatment
William´s test p-value * p<0.025, NS: Not significant.

Figure 37: Change from baseline (LSM and 95% CI) in the percentage of awake time per day spent in the OFF state based on on patient´s ON/OFF diary (observed case analysis)



Total hours of awake time spent in the ON state without troublesome dyskinesia.

The LSM differences from the placebo group at Endpoint in the total hours of awake time per day spent in the ON state without troublesome dyskinesia were 0.57 hours [95% C.I.: -0.08 hours to 1.22 hours] for the 20 mg/day KW-6002 group and 0.65 hours [0.01 hours to 1.28 hours] for the 40 mg/day KW-6002 group (See Table 75), equivalent to an increase in ON time without troublesome dyskinesia of 34 and 39 minutes respectively.

Table 75: Total hours of awake time per day spent in the ON state without troublesome dyskinesia based on patient´s ON/OFF diary-actual and change from baseline values (observed case analysis)

			Placebo n=118		20mg/day n=115		40mg/day n=124	
			Actual	Change	Actual	Change	Actual	Change
Baseline	Statistics	n	118		115		124	
		Mean	9.09		9.07		9.34	
		Std Dev	2.73		2.75		2.80	
		Minimum	1.5		1.3		1.4	
		Median	9.45		9.10		9.25	
		Maximum	15.3		15.0		16.5	
Endpoint	Statistics	n	118	118	115	115	124	124
		Mean	9.74	0.65	10.28	1.21	10.59	1.25
		Std Dev	3.37	2.49	3.47	2.36	3.04	2.80
		Minimum	0.0	-7.7	1.6	-5.3	2.0	-5.0
		Median	9.90	0.70	10.30	1.30	10.40	1.00
		Maximum	16.6	8.5	18.0	8.4	19.2	12.8
	ANCOVA	LS Mean	9.81	0.64	10.39	1.21	10.46	1.29
		95%C.I.		0.18, 1.11		0.74, 1.69		0.83, 1.75
	ANCOVA (vs. Placebo)	LS Mean				0.57		0.65
		95%C.I.				-0.08, 1.22		0.01, 1.28
		p-value				0.085		0.048
	Williams Test	Test Statistic, DF				-		2.01, 321
		p-value				-		0.026 NS

LSM and p-value are based on the main effects ANCOVA with terms for baseline, investigator and treatment
William´s test p-value * p<0.025, NS: Not significant.

UPDRS II and III in the ON state.

There was no difference in change in UPDRS 11 scores in the ON state from baseline to endpoint. For the UPDRS 111 there was a difference of 2 in favour of both istradefylline groups compared to placebo for reduction in UPDRS part 111 score from baseline (

Table 76).

Table 76: UPDRS Part III subscale score in the ON state-Actual and change from baseline (observed case analysis)

			Placebo n=118		20 mg/day n=115		40 mg/day n=124	
			Actual	Change	Actual	Change	Actual	Change
Day -1	Statistics	n	118		115		124	
		Mean	20.6		21.0		21.1	
		Std Dev	9.2		10.6		11.0	
		Minimum	1		2		0	
		Median	21.0		20.0		20.0	
		Maximum	49		61		53	
End-point	Statistics	n	118	118	115	115	124	124
		Mean	16.7	-3.9	14.9	-6.0	15.2	-5.9
		Std Dev	9.4	5.4	10.1	8.3	11.1	6.5
		Minimum	0	-24	0	-46	0	-29
		Median	18.0	-3.0	13.0	-4.0	14.0	-4.0
		Maximum	48	10	63	11	49	7
	ANCOVA	LS Mean	17.2	-3.7	15.2	-5.7	15.2	-5.7
		95%C.I.		-4.8, -2.6		-6.8, -4.6		-6.8, -4.6
	ANCOVA (vs. Placebo)	LS Mean				-2.0		-2.0
		95%C.I.				-3.5, -0.4		-3.5, -0.4
		p-value				0.013		0.012
	Williams Test	Test Statistic, DF				2.52, 321		2.56, 321
		p-value				0.006 *		0.006 *

LSM and p-value are based on the main effects ANCOVA with terms for baseline, investigator and treatment
William´s test p-value * p<0.025, NS: Not significant.

Ancillary analyses

For the change from Baseline in the total hours of awake time per day spent in the OFF state at Endpoint (observed case analysis) based on the FAS, stratified analyses by the following subgroup factors were performed: main demographic and baseline characteristics, i.e., age, gender, BMI, time since diagnosis, time since onset of motor complications, current smoker, Modified Hoehn and Yahr stage at Day -1 (OFF state), total hours of awake time per day spent in the OFF state at Baseline, percentage of awake time per day spent in the OFF state at Baseline, daily dosage of prior levodopa, and combinations of concomitant antiparkinsonian drugs. The summary statistics of the change in the total hours of awake time per day spent in the OFF state in each subgroup were calculated by treatment.

The p-value was less than 0.05 for the main effect of "Age" (p=0.002) and that of "BMI" (p=0.042) in the ANCOVA model without a treatment-by-subgroup interaction term. The increase in the "65 years and older" subset relative to the "younger than 65 years" subset was 0.78 hours [95% C.I.: 0.29 hours to 1.27 hours], indicating that the subjects aged less than 65 years experienced a smaller reduction in the total hours of awake time per day spent in the OFF state.

The increase in the "22 mg/m2 or more" subset relative to the "less than 22 mg/m2" subset was 0.51 hours [95% C.I.: 0.02 hours to 0.99 hours], indicating that the subjects with a BMI of less than 22 mg/m2 experienced a smaller reduction in the total hours of awake time per day spent in the OFF state.

Summary of main efficacy results

This summary should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 77: Summary of efficacy for trial 6002-0608

Title: Placebo-Controlled, Double-Blind, Parallel-Group, Fixed Dose Study of KW-6002 (Istradefylline) in the Treatment of Parkinson's Disease					
Study identifier	6002-0608				
Design	Phase 2b, randomised, double-blind, placebo-controlled, fixed-dose, parallel-group, multicenter trial.				
	Duration of main phase:		12 weeks		
	Duration of Run-in phase:		Not applicable		
	Duration of Extension phase:		Not applicable		
Hypothesis	Superiority				
Treatments groups	Istradefylline 20 mg		Istradefylline 20 mg once daily, 12 weeks, n=119 subjects		
	Istradefylline 40 mg		Istradefylline 40 mg once daily, 12 weeks, n=125 subjects		
	Placebo		Placebo once daily, 12 weeks, n=119 subjects		
Endpoints and definitions	Primary endpoint	OFF time per day	Change from baseline in total hours of awake time/day spent in the OFF state		
	Key secondary endpoint	ON time per day without troublesome dyskinesia	Change from baseline in total hours of ON time/day without troublesome dyskinesia		
Database lock	24 September 2008				
Results and Analysis					
Analysis description	**Primary Analysis**				
Analysis population and time point description	Full Analysis Set (FAS), week 12/Endpoint LOCF, Observed case, ANCOVA				
Descriptive statistics and estimate variability	Treatment group		Placebo	Istradefylline 20 mg	Istradefylline 40 mg
	Number of subjects analysed at Week 12/ Endpoint		118	115	124
	Change from baseline in total hours of awake time/day spent in the OFF state; Mean (SD)		-0.62 (2.17)	-1.36 (2.01)	-1.55 (2.66)
	Range (min to max)		-8.0 to 7.0	-7.1 to 3.0	-10.7 to 5.3
	Change from baseline in total hours of ON time/day without troublesome dyskinesia; Mean (SD)		0.65 (2.49)	1.21 (2.36)	1.25 (2.80)
	Range (min to max)		-7.7 to 8.5	-5.3 to 8.4	-5.0 to 12.8
Effect estimate per comparison	Primary endpoint (hours/day spent in the OFF state); difference from placebo in the LSM change from baseline		Comparison groups	Istradefylline 20 mg vs Placebo	Istradefylline 40 mg vs Placebo
			LSM change vs. placebo	-0.65	-0.92

		95% CI	-1.23, -0.07	-1.49, -0.35
		P-value	0.028	0.002
	Key secondary endpoint (hours/day spent ON without troublesome dyskinesia); difference from placebo in the LSM change from baseline	Comparison groups	Istradefylline 20 mg vs Placebo	Istradefylline 40 mg vs Placebo
		LSM change vs. placebo	0.57	0.65
		95% CI	-0.08, 1.22	0.01, 1.28
Notes	Primary endpoint: Change from baseline for istradefylline vs. placebo by Williams' test, where p<0.025 indicates statistical significance: p-value=0.013 (20mg), p-value<0.001 (40mg)			

Study 6002-009

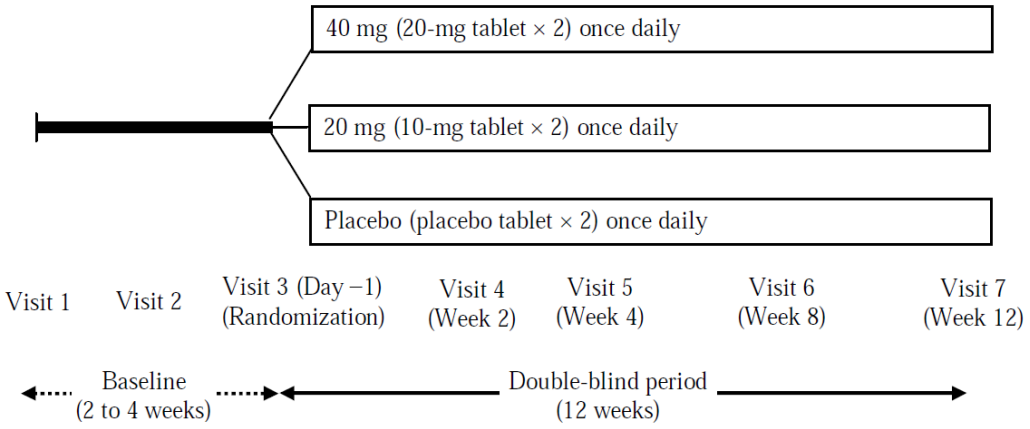
"Placebo-Controlled, Double-Blind, Parallel-Group, Confirmatory Comparative Study of KW-6002 in the Treatment of Parkinson's Disease (Phase 3 Study)"

Methods

Study design

This was a multicentre, placebo-controlled, randomized, DB, parallel-group comparative and confirmatory phase 3 study of KW-6002 administered at an oral dose of 20 or 40 mg once daily for 12 weeks in PD patients with motor complications on levodopa therapy

Figure 38: Study design of Study 6002-009



Study participants

Inclusion criteria

- Aged 20 years or more diagnosed with PD based on UKPDS Brain bank criteria for PD.
- PD in Stages 2-4 modified H&Y Scale while in an OFF state on the day of randomisation (D -1).
- Had responded to levodopa/DCI (carbidopa or benserazide) and had been treated for at least 1 year without interruption prior to randomisation, were taking at least 3 doses of levodopa/DCI per day with a daily dose of at least 300mg (excluding levodopa monotherapy) within 4 weeks prior to randomisation and with predictable end of dose wearing off.
- Successful completion of diary training.
- A total of at least 2 hour of OFF time per day recorded on at least 4 valid diaries completed during 7 days prior to randomisation after successfully completing diary training.
- No new anti PD drugs and a stable regiment of anti-PD drugs for at least 4 weeks prior to randomisation.
- No change in prescription of prophylactic treatment with domperidone within 14 days prior to Visit 3 (randomisation).

Main Exclusion criteria

- Treatment with CYP3A4 substrates or inhibitors or zonisamide within 2 weeks prior to randomisation.
- Treated with MAO inhibitors except selegiline within 3 months prior to randomisation.
- Treated with a drug that is a substrate of P-glycoprotein within 14 days prior to Visit 3 (day of randomisation).
- Treated within 3 months prior to randomisation with typical or atypical antipsychotics (6 months for depot formulations), centrally acting dopamine antagonists, reserpine and papaverine.
- Neurosurgical operation for PD (stereotactic surgery, or deep brain stimulation) at any time; or transcranial magnetic stimulation within 6 months prior to randomisation.
- Prior exposure to KW-6002 or exposure to any other IP within 4 months prior to randomisation.
- History of dementia or a score of ≤ 23 on the mini mental state examination conducted during the baseline period.
- History of psychotic illness, apart from psychiatric symptoms related to PD.

Treatments

Treatment with IP began on the day after visit 3 (Day-1):

- KW-6002 20mg/day: two KW-6002 10mg tablets administered once daily for 12 weeks.
- KW-6002 20mg/day: two KW-6002 20mg tablets administered once daily for 12 weeks.
- Placebo: two placebo tablets administered once daily for 12 weeks.

Objective

The objective of this study was to evaluate the efficacy of KW-6002, administered at an oral dose of 20 or 40 mg once daily for 12 weeks, in subjects with PD with motor complications on levodopa therapy based on the change in the total hours of awake time per day spent in the OFF state compared with placebo and to evaluate the safety of KW-6002 at 20- and 40 mg/day.

Outcomes/endpoints

Primary endpoint

Change in the total hours of awake time per day spent in the OFF state (total hours per day spent in the OFF state) assessed at the last assessment time at which the endpoint was evaluated

The total hours of awake time per day spent in the OFF state was defined as the number of 30-minute periods classified as "OFF" in the ON/OFF diary multiplied by 0.5 hours. At each assessment time, the following diaries were to be included in the evaluation of the total hours of awake time per day spent in the OFF state. The end of treatment visit date (or the ET date if the ET visit did not fall on the date of ET) was considered the date on which a scheduled visit took place following the last scheduled visit prior to ET.

- Patient's ON/OFF diaries during the acceptable time windows below within the seven days preceding each visit
    - Baseline period On or after the day following Visit 2
    - Week 2 + 4 to 16 days beginning from Visit 3 (randomisation)
    - Week 4 + 18 to 30 days beginning from Visit 3
    - Week 8 + 46 to 58 days beginning from Visit 3
    - Week 12 + 74 to 86 days beginning from Visit 3

The total hours of awake time per day spent in the OFF state were calculated from each of the diaries subject to evaluation, and the mean value for valid diaries was used as the total hours of awake time per day spent in the OFF state at a given assessment time. If less than four days of valid diaries were available for evaluation at a particular assessment time, the data for that assessment time was consider missing.

Secondary endpoints

The following secondary endpoints were determined at each post-dose assessment times. The primary assessment times for variable evaluated at multiple post-dose assessment times was the last assessment time at which the variable was evaluated (final evaluation) for each subject.

- Change in the percentage of awake time per day spent in the OFF state (% of awake time per day spent in the OFF state)
- Changes in the total hours of awake time per day spent in the ON state with or without dyskinesia and by degree of dyskinesia (total hours per day spent in the ON state)
- Changes in the percentage of awake time per day spent in the ON state with or without dyskinesia and by degree of dyskinesia (% of awake time per day spent in the ON state)
- Changes in the UPDRS Part I, Part II (ON and OFF states), Part III (ON state), and Part IV scores, and Parts I to III total score (ON state)
- CGI-I

- Change in the modified H&Y staging

Sample size

The required sample size was estimated to be 120 in each treatment group based on results from the Study 6002-US-005 comparing the 40mg dose with placebo and the phase 2b study (6002-608) which compared the 20mg and 40mg doses with placebo (both had similar inclusion/exclusion criteria to Study 6002-009).

The results of an ANCOVA analysis in Study 6002-US-005 showed that the LSM difference relative to placebo in the total hours of awake time per day spent in the OFF state was −1.1 hours for 40 mg/day KW-6002 with a SD of 2.5 hours. In Study 6002-0608, the results of an ANCOVA analysis showed that the LSM change from baseline relative to placebo in the total hours of awake time per day spent in the OFF state was estimated to be −0.7 hours for subjects treated with 20 mg/day KW-6002 and −0.9 hours for subjects treated with 40 mg/day KW-6002, with a standard deviation of error of 2.2 hours.

An effect size of 40% was selected for this study, given the above results and the expectation for 1-hour difference in efficacy (as measured by the reduction from baseline in the total hours of awake time per day spent in the OFF state) between KW-6002 20mg or 40 mg/day and placebo with an estimated standard deviation of 2.5 hours. The number of subjects required was determined to be 110 per group in order to confirm the efficacy of KW-6002 20mg or 40 mg/day relative to placebo at a one-sided significance level of 2.5% and a detection power of 80% by the Williams' test. To account for a small percentage of subjects who are expected to be excluded from the analysis set, a sample size of 120 subjects per group (360 subjects in three groups in total) was planned.

Randomisation

Randomization was achieved by use of the permuted block method to make allocation to subjects in blocks by investigational site. The treatment assignment manager randomly assigned subjects in each block to 3 groups: KW-6002 20- and 40 mg/day and placebo. The IP for each subject was sealed. The investigators assigned the lowest subject No. in the block available then to eligible subjects in the order they were randomized and prescribed the corresponding IP.

Blinding (masking)

After individual subjects completed their treatment, the IP manager at the investigative site checked and sealed all unused drug for collection by the Sponsor. The Sponsor collected the unused drug and maintained custody and control of the returned drug under seal; no un-blinding of the unused IP took place.

The treatment assignment manager verified that all emergency codes remained unbroken when the sealed allocation tables were opened. Further, the Sponsor received plasma drug concentration data from the contract laboratory for measuring drug concentrations after the allocation tables were unsealed. Based on the above, the blinding of IP was ensured during the conduct of the study.

Statistical methods

The SAP was to be finalised before data lock. The statistical analysis plan was completed on 19 April 2011

<u>Primary endpoint</u>

An ANCOVA model was to be used for the principal analysis of the total hours of awake time per day spent in the OFF state (primary endpoint). An ANCOVA model using treatment group as a factor and baseline values and study centre as covariates was to be fitted to the change from baseline in the total hours of awake time per day spent in the OFF state at each post-baseline assessment or final evaluation. The LSM and 95% CI was to be calculated for each treatment group to estimate the change in total hours wake per day spent in the OFF state. The LSM and 95% CI were also be calculated for each istradefylline 20mg and 40mg per day.

To determine efficacy of istradefylline relative to placebo at the final evaluation, 20mg/day and 40mg/day were to be compared to placebo using the William's test at a one sided significance level of 2.5%.

The study centres used in the ANCOVA model were to be defined by investigator. If the number of subjects were so small that some centres need to be combined, this was to be determined through blinded review before data lock.

Secondary endpoints

An ANCOVA model using the change from baseline as a response variable and baseline value and study center as covariates was to be fitted to the percentage of awake time per day spent in the OFF state, total hours and percentage of awake time per day spent in the ON state by state of dyskinesia, and UPDRS scores. The LSM and its 95% CI were to be calculated for each treatment group. In addition, the LSM difference from placebo and its 95% CI were to be calculated for 20 mg/day and 40 mg/day KW-6002.

The frequency of CGI-I evaluation at the final evaluation were to be determined by treatment group. In addition, the percentages of subjects in the "much improved" or better category ("very much improved" + "much improved") and in the "minimally improved" or better category ("much improved" or better + "minimally improved") were to be calculated by treatment group.

The change from baseline in the modified Hoehn and Yahr staging score at Week 12/ET in each treatment group was to be summarised using shift tables.

Handling of missing data

1. If data at Week 12 or ET are missing in the analyses of the primary and secondary variables, the LOCF approach will be used, i.e., the last available post-baseline value will be used as the final-evaluation data.
2. In patient's ON/OFF diaries, 30-minute periods with invalid entries will be handled as missing and will not be included in the analysis (observed-case analysis). The stability of the observed-case analysis will be assessed by a worst-case analysis, in which 30-minute periods with missing entries are to be substituted with OFF, and those periods with multiple entries are to be substituted with the worst-case value to be selected from one of the following states in the order of priority given: asleep < ON state without dyskinesia < ON state with non-troublesome dyskinesia < ON state with troublesome dyskinesia < OFF.
3. When calculating the UPDRS scores by part, if one or more subscale components of a part are missing, then the score for that part is to be handled as missing. Any missing values at the final evaluation are to be substituted with the last available post-baseline values using the LOCF approach before determining the score by part.

Multiplicity

The principal analysis in this clinical study for the primary variable at the final evaluation in the FAS will be Williams' test (one-sided significance level of 2.5%). Overall, the type I error probabilities for the comparisons of 20 mg/day and 40 mg/day KW-6002 versus placebo in the principal analysis will kept

fixed for the overall level of significance. Other analyses will be considered secondary analyses complementary to the principal analysis and the issue of multiplicity will not be a consideration.

Results

Participant flow

398 patients were screened of whom 373 were randomised. Of the 25 screen failure, 5 failed to meet inclusion criteria, and 11 met exclusion criteria, and 8 requested withdrawal.

Table 78: Distribution of participants

	Placebo	Istradefylline 20mg/day	Istradefylline 40mg/day
Randomised	126	123	124
Exposed to IP	126	123	124
Completed week 12	109 (86.5%)	111 (90.2%)	115 (92.7%)
Completion			
Week 2	124 (98.4%)	121 (98.4%)	123 (99.2%)
Week 4	119 (94.4%)	119 (96.7%)	120 (96.8%)
Week 8	112 (88.9%)	113 (91.9%)	117 (94.4%)
Week 12	109 (86.5%)	111 (90.2%)	115 (92.7%)
Early withdrawal by cause			
TEAE	6 (4.8%)	5 (4.1%)	6 (4.8%)
Protocol violation	1 (0.8%)	0	0
Drug non-compliance	2 (1.6%)	0	0
Subject request	6 (4.8%)	6 (4.9%)	3 (2.4%)
Investigator’s discretion	2 (1.6%)	1 (0.8%)	0

A total of 38 (10.2%) withdrew prematurely from the study: 17 (13.5%) in the placebo group, 12 (9.8%) in the Istradefylline 20 mg/day, and 9 (7.3%) in the Istradefylline 40 mg/day group. The reasons for withdrawal were: TEAE, (placebo, 6 (4.8%); istradefylline 20 mg/day, 5 (4.1%), istradefylline 40 mg/day, 6 (4.8%); protocol violation, 1 (0.8%) placebo; drug non-compliance, 2 1.6%placebo; subject’s own request, placebo, 6 (4.8%); Istradefylline 20 mg/day, 6 (4.9%); Istradefylline 40 mg/day, 3 (2.4%); and investigator’s discretion placebo, 2 (1.6%); Istradefylline 20 mg/day, 1 (0.8%). No bias was noted in the time of withdrawal among groups. The rest of the 335 subjects (89.8%) completed the 12-week treatment: placebo group, 109 (86.5%); Istradefylline 20 mg/day, 111 (90.2%); and Istradefylline 40 mg/day, 115 (92.7%).

Recruitment

Study locations: 44 centers in Japan

Study period: study initiation was on 21 July 2009 when the first patient was consented and study completion was on 21 February 2011 (date of completion of the protocol specified examination for the last participant)

Conduct of the study

Protocol amendments

There were 3 protocol amendments (ver.1.1 1 July 2009; ver1.2 21 May 2010 and ver1.3 17 August 2010) with no major changes. No changes were made to the SAP.

Protocol Deviation

In this study, significant protocol deviations were reported in one subject who was enrolled in the study despite failing to meet the inclusion criteria; seven subjects who failed to comply with drug regimen (an overall drug compliance rate of less than 70% during the study in those who withdrew; or an overall drug compliance rate of less than 70% during the study or between "Weeks 8 and 12" in those who completed the study); and 10 subjects who received a prohibited concomitant therapy.

Baseline data

In the FAS, population (n=366), the placebo, KW-6002 20 mg/day, and 40 mg/day of KW-6002 groups, respectively, had: a mean age of 65.8 ± 8.6 (SD), 66.1 ± 8.6, and 65.7 ± 9.0 years; and 65 (52.8%), 80 (66.7%), and 59 (48.0%) were females; a history of PD for 8.0 ± 4.5, 7.3 ± 4.2, and 7.7 ± 4.5 years; and a history of motor complications for 3.4 ± 3.5, 3.2 ± 2.8, and 3.3 ± 3.0 years.

The H&Y stage (ON) was "2 to 3" in about 90% of the subjects in each group, while the modifed H&Y stage (OFF) was "3 to 4" in about 80% of the subjects in each group.

The total hours of awake time per day spent in the OFF state was, respectively, 6.31 ± 2.47, 6.55 ± 2.72, and 5.97 ± 2.45 hours, with the total percentage of the same being 38.91 ± 14.80%, 40.59 ± 16.19%, and 36.92 ± 15.10%, according to the ON/OFF diaries.

Table 79: Demographic and other baseline characteristics

	Placebo N =123	KW20 mg/day N = 120	KW40 mg/day N = 123
Age Mean (SD) Median (Min, Max) ≥ 85 years	 65.8 (8.6) 66 (48, 83) 69 (56.1)	 66.1 (8.6) 67 (42, 84) 72 (60)	 65.7 (9) 68 (33, 82) 76 (61.8)
Gender F M	 65 (52.8%) 58 (47.2%)	 80 (66.7%) 40 (33.3%)	 59 (48%) 64 (52%)
Duration of PD (year) Mean (SD) Median (Min, Max) ≥ 10 years	 .58 (4.453) 7.540 (1.14, 26.02) 36 (29.3%)	 7.301 (4.206) 6.375 (0.63, 20.02) 30 (25%)	 7.73 (4.547) 7.23 (0.66, 22.8) 31 (25.2%)
Duration of motor complications (years) Mean (SD) Median (Min, Max) ≥ 4 years	 3.432 (3.47) 2.5 (0.05, 22.02) 36 (29.3%)	 3.183 (2.759) 2.46 (0.04, 13.14) 33 (27.5%)	 3.258 (3.009) 2.51 (0.04, 18.14) 34 (27.6%)
Hoehn &Yahr ON 0 1 1.5 2 2.5 3 4	 0 9 (7.3%) 2 (1.6%) 41 (33.3%) 34 (27.6%) 32 (26%) 5 (4.1%)	 2 (1.7%) 6 (5%) 1 (0.8%) 44 (36.7%) 33 (27.5%) 26 (21.7%) 8 (6.7%)	 0 5 94.1%) 8 (6.5%) 43 (35%) 30 (24.4%) 32 (26%) 5 (4.1%)
Hoehn & Yahr OFF 2 2.5 3 4	 7 (5.7%) 16 (13%) 52 (42.3%) 48 (39%)	 9 (7.5%) 17 (14.2%) 44 (36.7%) 50 (41.7%)	 10 (8.1%) 12 (9.8%) 49 (39.8%) 52 (42.3%)
Total hours awake OFF			

Mean (SD) Median (Min, Max) ≥ 6 hours	6.31 (2.47) 6.1 (1.9, 13.1) 65 (52.8%)	6.55 (2.72) 6.2 (2, 14.2) 62 (51.7%)	5.97 (2.45) 5.8 (2, 13.8) 56 (45.5%)

The levodopa dose was, respectively, 425.4 ± 146.4, 430.8 ± 156.5, and 420.5 ± 131.8 mg/day.

Subjects on a dopamine agonist administered as a concomitant antiparkinsonian drug accounted for 91.1%, 85.8%, and 83.7%, respectively. Other commonly used concomitant drugs were selegiline (46.3%, 43.3%, and 61.0%) and entacapone (42.3%, 52.5%, and 55.3%). Higher proportions of those in the Istradefylline 40mg/day arm were treated with selegiline or entacapone compared to the placebo arm. On the other hand a higher proportion of those in the placebo arm were treated with pramipexole compared to both Istradefylline groups (Table 80).

Table 80: Baseline PD treatment

Baseline PD treatment			
	Placebo N =123	KW20 mg/day N = 120	KW40 mg/day N = 123
Levodopa dose (mg/day) Mean (SD) Median (min, max)	 425.4 (146.4) 400 (300, 1200)	 430.8 (156.5) 400 (300, 1200)	 420.5 (131.8) 400 (300, 900)
Dopamine agonist	112 (91.1%)	103 (85.8%)	103 (83.7%)
Pramipexole	77 (62.6%)	65 (54.2%)	57 (46.3%)
Selegiline	57 (46.3%)	52 (43.3%	75 (61%)
Entacapone	52 (42.3%)	63 (52.5%)	68 (55.3%)
Amantadine	49 (39.8%)	41 (34.2%)	44 (35.8%)
Combination of concomitant PD drugs Levodopa Levodopa +DA Levodopa +DA +SEl/ENT/ZNS Levodopa+DA SEl/ENT/ZNS+ AMA Other	 2 (1.6%) 19 (15.4%) 49 (39.8%) 33 (26.8%) 20 (16.3%)	 1 (0.8%) 15 (12.5%) 56 (46.7%) 24 (20%) 24 (20%)	 3 (2.4%) 13 (10.6%) 52 (42.3%) 31 (25.2%) 14 (19.5%)

Numbers analysed

In this study, 398 subjects were screened (i.e., gave informed consent). Of these subjects, 25 were not randomized and dropped out during the baseline period for the following reasons: subject's own request, 8; failure to meet the inclusion criteria, 5; meeting the exclusion criteria, 11; and other, 1.

Analyses of the primary and secondary variables were conducted principally on the "FAS" consisting of subjects who were randomised, received at least one dose of IP and submitted at least 4 days of valid diaries subject to evaluation at any of the post-baseline assessment times.

The "PPS" consisting of subjects in the FAS who met all of the inclusion criteria and none of the exclusion criteria specified in the protocol, completed the 12-week DB treatment, achieved a drug compliance rate of 70% or more during both the entire treatment period and the interval from "Weeks 8 to 12" and had no major protocol deviations. The PP set was analysed to confirm the stability of the principal analysis of the primary variable.

Table 81: Analysis set by treatment arm

Analysis sets by treatment arm			
	Placebo	KW 20mg/day	KW40 mg/day
Randomised	126	123	124
Full analysis set	123 (97.6%)	120 (97.6%)	123 (99.2%)
Reasons for exclusion			
No valid diaries of ≥ 4 days	3	3	1
Completion			
Week 2	124 (98.4%)	121 (98.4%)	123 (99.2%)
Week 4	119 (94.4%)	119 (96.7%)	120 (96.8%)
Week 8	112 (88.9%)	113 (91.9%)	117 (94.4%)
Week 12	109 (86.5%)	111 (90.2%)	115 (92.7%)
Per protocol population	107 (84.9%)	109 (88.6%)	109 (87.9%)

18 patients had 'significant' protocol deviations, 8 in the placebo group, 3 in the KW20mg/day group and 7 in the KW40 mg/day group. 16 subjects were excluded from the PP set from the placebo arm (1 violation of study inclusion/exclusion criteria; 14 failure to complete 12W treatment; 2 drug compliance rate <70% and 4 major protocol deviation) , 11 from the KW20mg/day arm (9 failure to complete 12W treatment; 1 drug compliance rate <70% and 1 major protocol deviation) and 14 from the KW 40mg/day arm (8 failure to complete 12W treatment; 1 drug compliance rate <70% and 5 major protocol deviation)).

Outcomes and estimation

Primary endpoint

For the total hours of awake time per day spent in the OFF state, the LSM change from baseline (95% CI) at the final evaluation estimated based on the ANCOVA model (observed case analysis in the FAS) was −0.23 h (−0.62 to 0.16 h) for the placebo group, −0.99 h (−1.38 to −0.60 h) for the 20 mg/day of KW-6002 group, and −0.96 h (−1.35 to −0.58 h) for the 40 mg/day of KW-6002 group. The LSM difference from the placebo group (95% CI) at the final evaluation was −0.76 h (−1.30 to −0.22 h) for the 20 mg/day of KW-6002 group and −0.74 h (−1.27 to −0.20 h) for the 40 mg/day of KW-6002 group. The results of Williams' test showed that, compared with placebo, 20 mg and 40 mg of KW-6002 significantly (p = 0.003 for both groups) reduced the total hours per day spent in the OFF state. Any missing data on the primary or secondary variable at Week 12 or ET was substituted with the last available post-baseline data using the LOCF approach. A worst case analysis in the FAS produced similar results. There is little separation between the Istradefylline 20mg and 40mg in terms of decrease in OFF time whilst awake.

Table 82: Total amount of awake time per day spent in the OFF state (observed case analysis)

	Placebo	KW 20mg/day	KW40 mg/day
Baseline	N = 123	N = 120	N =123
Mean (SD)	6.31 (2.47)	6.55 (2.72)	5.97 (2.45)
Median (Min, max)	6.1 (1.9, 13.1)	6.2 (2, 14.2)	5.8 (2, 13.8)
Endpoint			
Mean (SD)	6.03 (3.05)	5.46 (2.93)	5 (2.84)
Median (Min, max)	5.9 (0, 14.8)	5.6 (0, 14.1)	5 (0, 12.1)

ANCOVA LSM change (95%CI)	-0.23 (-0.62, 0.16)	-0.99 (-1.38, -0.6)	-0.96 (-1.35, -0.58)
ANCOVA LSM change vs. Placebo (95% CI)		-0.76 (-1.3, -0.22)	-0.74 (-1.27, -0.2)
Williams test P value		0.003	0.003

ANCOVA: a model with treatment group as a factor and baseline value and investigative site as covariates
Williams' test p-value*: $p < 0.025$

Figure 39: Total Hours of Awake Time per Day Spent in the OFF state (LSM and 95% CI) (observed case analysis)

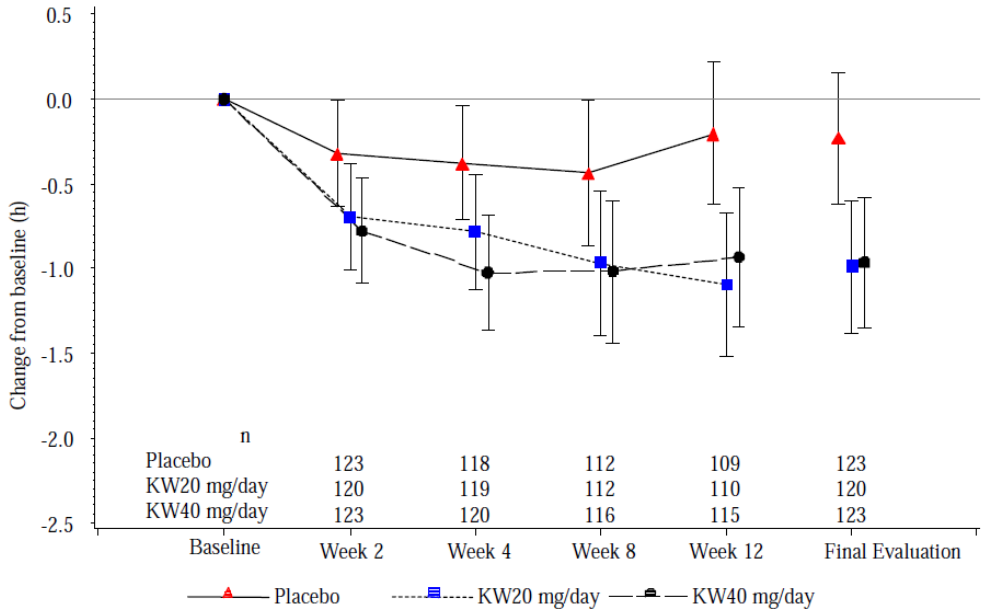


Table 83: Total amount of awake time per day spent in the OFF state (worst case analysis)

	Placebo	KW 20mg/day	KW40 mg/day
Baseline Mean (SD) Median (Min, max)	N = 123 6.31 (2.47) 6.1 (1.9, 13.1)	N = 120 6.56 (2.72) 6.2 (2, 14.2)	N =123 5.97 (2.45) 5.8 (2, 13.8)
Endpoint Mean (SD) Median (Min, max)	 6.03 (3.05) 5.9 (0, 14.8)	 5.47 (2.94) 5.6 (0, 14.1)	 5.01 (2.85) 5 (0, 12.1)
ANCOVA LSM change (95%CI)	-0.22 (-0.61, 0.16)	-0.99 (-1.38, -0.59)	-0.96 (-1.34, -0.57)
ANCOVA LSM change v placebo (95% CI)		-0.76 (-1.31, -0.22)	-0.73 (-1.27, -0.19)
Williams test P value		0.003	0.003

LSM and p-value are based on the main effects ANCOVA with terms for Baseline, Investigator and treatment
Williams' test p-value *: $p<0.025$; NS: Not Significant

Secondary endpoints

Percentage of awake time per day spent in the OFF state

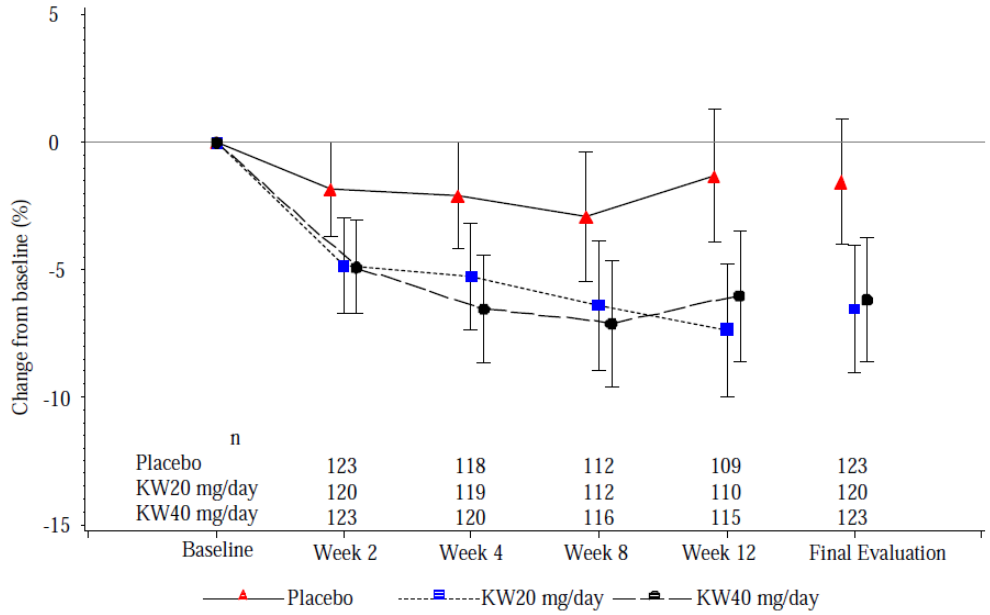
For the % of awake time per day spent in the OFF state, the LSM change from baseline (95% CI) at the final evaluation estimated based on the ANCOVA model was −1.55% (−3.98% to 0.88%) for the placebo group, −6.55% (−9.02% to −4.08%) for the 20 mg/day of KW-6002 group, and −6.17% (−8.59% to −3.74%) for the 40 mg/day of KW-6002 group. The LSM difference from the placebo group (95% CI) at the final evaluation was −4.99% (−8.39% to −1.60%) for the 20 mg/day of KW-6002 group and −4.61% (−7.98% to −1.25%) for the 40 mg/day of KW-6002 group. The results of Williams' test showed that, compared with placebo, 20 mg and 40 mg of KW-6002 significantly (p = 0.002 and 0.003, respectively) reduced the % of awake time per day spent in the OFF state (

Table 84).

Table 84: Percentage of awake time per day spent in the OFF state (observed case analysis)

	Placebo	KW 20mg/day	KW40 mg/day
Baseline Mean (SD) Median (Min, max)	N = 123 38.91 (14.8) 37.7 (11.6, 85.3)	N = 120 40.59 (16.19) 38.65 (12.5), 90.8)	N =123 36.92 (15.1) 36.2 (10.7, 83.7)
Endpoint Mean (SD) Median (Min, max)	 37.13 (18.4) 36.4 (0, 92.2)	 33.54 (17.88) 33.85 (0, 87.2)	 30.85 (17.38) 30.9 (0, 71.3)
ANCOVA LSM change (95%CI)	-1.55 (-3.98, 0.88)	-6.55 (-9.02, -4.08)	-6.17 (-8.59, -3.74)
ANCOVA LSM change v placebo (95% CI)		-4.99 (-8.39, -1.6)	-4.61 (-7.98, -1.25
Williams test P value		0.002	0.003

Figure 40: Percentage of Awake Time per Day Spent in the OFF state (LSM and 95% CI) (observed case analysis)



The 30-minute periods with an invalid entry in the ON/OFF diaries were handled as missing data and not included in the observed-case analysis, the principal analysis. The stability of the observed-case analysis was assessed by a worst-case analysis, in which 30-minute periods with missing entries were substituted with OFF, and those periods with multiple entries were substituted with the worst-case value to be

selected from one of the following states in the order of priority given: "asleep" < "ON state without dyskinesia" < "ON state with non-troublesome dyskinesia" < "ON state with troublesome dyskinesia" < "OFF". Both the observed- and worst-case analyses gave practically the same results, indicating minimal impact from the 30-minute periods with an invalid entry.

Total Hours of Awake Time per Day Spent in the ON State (without Dyskinesia)

The LSM change from baseline at endpoint in the placebo group was 0.28h (-0.16, 0.73) compared to 0.90 (0.44, 1.35 for Istradefylline 20mg and 0.85 (0.41, 1.3) for Istradefylline 40mg. The LSM difference from the placebo group (95% CI) at the final evaluation was 0.61 h (−0.01 to 1.24 h) in the 20 mg/day of KW-6002 group and 0.57 h (−0.05 to 1.19 h) in the 40 mg/day of KW-6002 group.

Total Hours of Awake Time per Day Spent in the ON State (without Troublesome Dyskinesia)

For the total hours of awake time per day spent in the ON state (without troublesome dyskinesia), the LSM change from baseline (95% CI) at the final evaluation estimated based on the ANCOVA model was 0.26 h (−0.17 to 0.70 h) for the placebo group, 1.09 h (0.65 to 1.54 h) for the 20 mg/day of KW-6002 group, and 1.08 h (0.64 to 1.51 h) for the 40 mg/day of KW-6002 group. The LSM difference from the placebo group (95% CI) at the final evaluation was 0.83 h (0.22 to 1.44 h) in the 20 mg/day of KW-6002 group and 0.81 h (0.21 to 1.42 h) in the 40 mg/day of KW-6002 group.

UPDRS part III (ON) score

For the UPDRS part III (ON) score, the LSM change from baseline at the final evaluation by treatment group, as estimated based on the ANCOVA model, was −2.8 in the placebo group, −3.7 in the 20 mg/day of KW-6002 group, and −4.9 in the 40 mg/day of KW-6002 group.

The LSM difference from the placebo group (95% CI) at the final evaluation was −0.9 (−2.3 to 0.4) in the 20 mg/day of KW-6002 group and −2.0 (−3.4 to −0.7) in the 40 mg/day of KW-6002 group.

Clinical Global Impression Global Improvement

The percentage of subjects with a score rated "2. much improved" or better at the final evaluation was 10.7% (13/122 subjects) in the placebo group, 20.8% (25/120) in the 20 mg/day of KW-6002 group, and 28.7% (35/122) in the 40 mg/day of KW-6002 group, and the percentage of subjects with a score rated "3. minimally improved" or better was, respectively, 36.9% (45/122), 57.5% (69/120), and 60.7% (74/122), indicating a high percentage of improvement relative to placebo.

Table 85: CGI-I frequencies at final evaluation (FAS population)

	Placebo n = 123	KW20 mg/day n = 120	KW40 mg/day n = 123
1.Very much improved	0(0.0%)	5(4.2%)	3(2.5%)
2.Much improved	13(10.7%)	20(16.7%)	32(26.2%)
3.Minimally improved	32(26.2%)	44(36.7%)	39(32.0%)
4.No change	69(56.6%)	40(33.3%)	42(34.4%)
5.Minimally worse	8(6.6%)	8(6.7%)	5(4.1%)
6.Much worse	0(0.0%)	2(1.7%)	1(0.8%)
7.Very much worse	0(0.0%)	1(0.8%)	0(0.0%)
Total	122(100%)	120(100%)	122(100%)
Wilcoxon two-sample test vs. placebo		z = -2.796 p = 0.005	z = 3.980 p < .001
2. "Much improved" or better	13(10.7%)	25(20.8%)	35(28.7%)
3. "Minimally improved" or better	45(36.9%)	69(57.5%)	74(60.7%)

Ancillary analyses

A number of subgroup analyses were performed. However, as in the case of secondary endpoints, these analyses should be interpreted with caution as they will not be sufficiently powered, and multiplicity of testing has not been considered here. A stratified analysis of the change from baseline at the final evaluation in the total hours of awake time per day spent in the OFF state (observed-case analysis) was conducted on the FAS with a model based on the ANCOVA model with the addition of major subject characteristics.

Summary of main efficacy results

This summary should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 86 Summary of efficacy for trial 6002-009

Title: Placebo-Controlled, Double-Blind, Parallel-Group, Confirmatory Comparative Study of KW-6002 in the Treatment of Parkinson's Disease			
Study identifier	6002-009		
Design	Phase 3, randomised, double-blind, placebo-controlled, fixed-dose, parallel-group, multicenter trial.		
	Duration of main phase: Duration of Run-in phase: Duration of Extension phase:		12 weeks Not applicable Not applicable
Hypothesis	Superiority		
Treatments groups	Istradefylline 20 mg		Istradefylline 20 mg once daily, 12 weeks, n=123 subjects
	Istradefylline 40 mg		Istradefylline 40 mg once daily, 12 weeks, n=124 subjects
	Placebo		Placebo once daily, 12 weeks, n=126 subjects
Endpoints and definitions	Primary endpoint	OFF time per day	Change from baseline in total hours of awake time/day spent in the OFF state

	Key secondary endpoint	ON time per day without troublesome dyskinesia	Change from baseline in total hours of ON time/day without troublesome dyskinesia		
Database lock	19 April 2011				
Results and Analysis					
Analysis description	**Primary Analysis**				
Analysis population and time point description	Full Analysis Set (FAS), week 12/Endpoint LOCF, Observed case, ANCOVA				
Descriptive statistics and estimate variability	Treatment group		Placebo	Istradefylline 20 mg	Istradefylline 40 mg
	Number of subjects analysed at Week 12/ Endpoint		123	120	123
	Change from baseline in total hours of awake time/day spent in the OFF state; Mean (SD)		-0.28 (2.32)	-1.10 (1.94)	-0.98 (2.13)
	Range (min to max)		-8.2 to 5.4	-9.0 to 3.8	-6.8 to 5.1
	Change from baseline in total hours of ON time/day without troublesome dyskinesia; Mean (SD)		0.26 (2.42)	1.17 (2.52)	1.01 (2.51)
	Range (min to max)		-6.6 to 7.7	-9.2 to 9.5	-6.4 to 9.3
Effect estimate per comparison	Primary endpoint (hours/day spent in the OFF state); difference from placebo in the LSM change from baseline		Comparison groups	Istradefylline 20 mg vs Placebo	Istradefylline 40 mg vs Placebo
			LSM change vs. placebo	-0.76	-0.74
			95% CI	-1.30, -0.22	-1.27, -0.20
			P-value	0.006	0.008
	Key secondary endpoint (hours/day spent ON without troublesome dyskinesia); difference from placebo in the LSM change from baseline		Comparison groups	Istradefylline 20 mg vs Placebo	Istradefylline 40 mg vs Placebo
			LSM change vs. placebo	0.83	0.81
			95% CI	0.22, 1.44	0.21, 1.42
Notes	Primary endpoint: Change from baseline for istradefylline vs. placebo by Williams' test, where $p<0.025$ indicates statistical significance: p-value=0.003 (20mg), p-value=0.003 (40mg)				

Study 6002-014

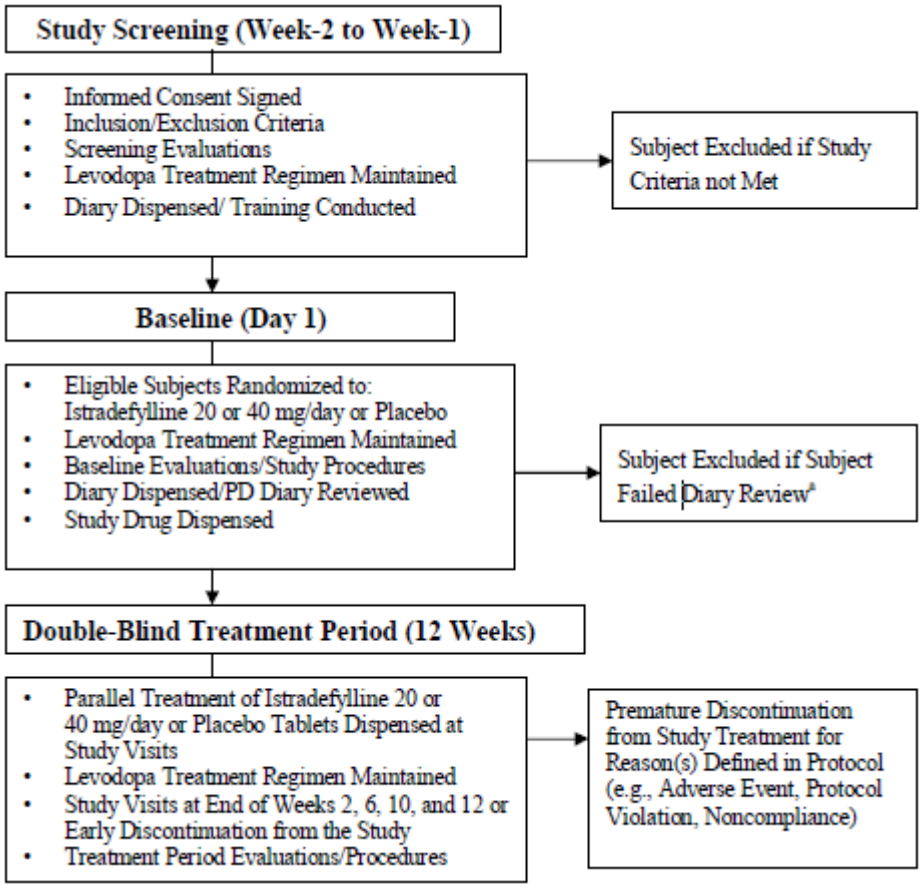
"A Phase 3, 12-week, Double-blind, Placebo-controlled, Randomized, Multicenter Study to Evaluate the Efficacy of Oral Istradefylline 20 and 40 mg/day as Treatment for Subjects with Moderate to Severe Parkinson's Disease"

Methods

Study design

This was a Phase 3, 12-week, DB, placebo-controlled, randomized, multicentre study to evaluate the efficacy and safety of istradefylline 20 and 40 mg/d compared to placebo in approximately 609 subjects with moderate to severe PD with motor fluctuations and dyskinesia on levodopa combination (L/C or benserazide/levodopa) therapy.

Figure 41: Study plan of Study 6002-014



a: Successfully completed diary training and a practice diary prior to Week -1 and be confirmed as having passed the training at Week -1, completed diary concordance testing and have achieved concordance of 80% with respect to both ON and OFF periods at the Week -1 visit, and successfully completed three valid ON/OFF patient diaries on the three consecutive days immediately prior to the baseline visit.

Study participants

Inclusion criteria

- Aged 30 years or over with a diagnosis of idiopathic PD based on UKPDRS Brain Bank Diagnostic Criteria, documented end-of dose-wearing off and levodopa induced dyskinesia.
- Minimum H&Y Scale stage classification of between 2 to 4 inclusive in the ON state. Note in the other pivotal study the inclusion criteria was a similar range of H&Y scores in the OFF state.

- Levodopa therapy for at least 1 year with continuing benefit (assessed at baseline visit) and stable dopaminergic regiment for at least 4 weeks immediately prior to randomisation.
- Currently taking L/C or L / benserazide with a total daily dose of a least 400mg, plus clinically effective dose of at least 1 adjunctive medication approved to treat PD.
- Successful completion of diary training and 3 valid ON/OFF diaries on 3 consecutive days immediately prior to baseline visit.

Main Exclusion criteria

- Atypical or secondary parkinsonism
- Currently treated with apomorphine and/or dopamine receptor antagonists or direct gastrointestinal levodopa infusion; treated with anticholinergic medications or amantadine alone
- History of a psychotic illness
- Previous neurosurgical procedure for PD
- Treated within 30 days before baseline with any investigational agent
- Previous istradefylline treatment of or currently receiving another A2a antagonist
- Taking potent CYP3A4 inhibitors or inducers

Treatments

All subjects were treated once daily with oral tablets of istradefylline 20 mg/d, 40 mg/d, or matching placebo tablets for 12 weeks in a 1:1:1 ratio. Patients continued with their established PD therapy throughout the trial PD therapy (subject to inclusion and exclusion criteria).

The treatments are similar to those evaluated in a number of the other pivotal studies. The dose selection has been justified by the applicant on the basis of previous receptor based

Objectives

The primary objective of the study was to establish the efficacy of istradefylline 20 and 40 mg/day (mg/d) for reducing the total hours of OFF time/day in moderate to severe PD patients with motor fluctuations and dyskinesia on levodopa combination therapy (levodopa/carbidopa or benserazide/levodopa).

The secondary efficacy objectives were the change from screening/baseline (as appropriate) in:

- Total hours of ON time/day without troublesome dyskinesia (key secondary objective).
- UPDRS Motor Examination Score (Part III); ADL score (Part II); and Mentation, Behaviour, and Mood (Part I).
- Total UPDRS (Parts I+II+III).
- PGI-I scale.
- Sleep time in hours per day based upon 24-hour diaries.
- The percentage of awake time/day spent in the OFF state.
- The percentage of ON time/day without troublesome dyskinesia.
- Total hours of ON time and percentage of ON time/day (without dyskinesia, with dyskinesia, with non-troublesome dyskinesia, and with troublesome dyskinesia).
- Montreal Cognitive Assessment (MoCA).

Outcomes/endpoints

Primary efficacy endpoint

The primary efficacy endpoint for this trial was the difference in the change from baseline to Week 12 in total hours OFF time between the 40 mg/d treatment group and the placebo group assessed from three 24 ON/OFF patient diaries completed on 3 consecutive days immediately prior to each scheduled visit. The primary efficacy analysis was based on the observed case analysis with the ITT analysis set. The difference in the change from baseline to Week 12 in total hours OFF time between the 20 mg/d treatment group and the placebo group was also examined.

Secondary Efficacy Endpoints

Total hours ON time without troublesome dyskinesia was considered the key secondary endpoint in this study.

Other secondary efficacy variables included the change from screening/baseline (as appropriate) in:

- Total hours of ON time/day without troublesome dyskinesia (key secondary objective).
- UPDRS Motor Examination (Part III).
- ADL score (Part II).
- Mentation, Behaviour, and Mood (Part I).
- Total UPDRS (Parts I, II, and III).
- PGI-I scale.
- Sleep time in hours per day based upon 24-hour diaries.
- Percentage of awake time spent in the OFF state.
- Percentage of ON time/day without troublesome dyskinesia.
- Total hours and percentage of ON time/day (without dyskinesia, with dyskinesia, with non-troublesome dyskinesia, and with troublesome dyskinesia).
- MoCA.
- BDI.

Subgroup analyses

Several subgroup analyses were conducted on the primary endpoint. An MMRM model with baseline value as a covariate, fixed effect terms in the model for site, treatment group, week, and treatment-by-week interaction, and an unstructured covariance matrix was run by region (i.e., North America v Rest of World) on the primary endpoint.

For subgroup analysis similar MMRM models used the following subject characteristics:

- Age (<65 v ≥65 years)
- Sex (male v female)
- Body Mass Index (< overall median value v ≥ overall median value)
- Duration of Parkinson's Disease (<1 v 1 to 3 v 4 to 7 v ≥8 years)
- Duration of Motor Complications (< overall median value v ≥ overall median value)
- Current Smoker Status (yes versus no)
- H&Y Scale Score (2 to 2.5 v 3 v 4)
- Total Hours Awake Time Spent in the OFF State at baseline (< overall median value v ≥ overall median value)
- Percent Hours Awake Time Spent in the OFF State at baseline (< overall median value v ≥ overall median value)
- Time Since Initiation of Levodopa (< overall median value v ≥ overall median value)

- Total Daily Levodopa Dose (400 to <600 v 600 to <800 v 800 to <1000 v ≥1000 mg)
- Concomitant use of MAO-B inhibitors (Yes v No)
- Concomitant use of dopamine agonists (Yes v No).
- Concomitant use of COMT inhibitors (Yes v No).

Sample size

To achieve 90% power for a MMRM with 4 post-baseline measurements, the correlation coefficient between total hours OFF time at baseline and change from baseline in total hours within the same subject was approximately 0.3834 based on previous istradefylline studies.

This same correlation was assumed for adjacent post-baseline values. The effect size considered was a change-from-baseline reduction in OFF time of 48 minutes (0.80 of an hour) based on a placebo change from baseline reduction of 0.7 hours and an istradefylline change from baseline reduction of 1.5 hours. The within-subject standard deviation for each treatment group was 2.7 hours. All of these assumptions were based on previous istradefylline studies.

Under these assumptions, a sample size of 185 subjects for each treatment group provided 90% power to detect a difference with an effect size of 0.8 hours (48 minutes) between the 40 mg treatment group and the placebo treatment group at the one-sided 2.5% significance level. The test used was based on an MMRM with 4 post-baseline measurements with the standard deviation within subjects of 2.7 hours. This power calculation extended to a 3-arm MMRM assuming the istradefylline 20 mg/d group had a change from baseline value between that of the placebo and istradefylline 40 mg/d groups. The proposed sample size per treatment group was increased by 9.5% (203 per treatment group) to account for trial drop out.

Randomisation

On Day 1 an Interactive Response Technology (IRT) was used to randomly assign each subject to a treatment assignment number generated by the IRT. The system was built according to Sponsor specifications and tested against those specifications prior to subject enrolment. The randomization schedule was stored with the IRT vendor.

Blinding (masking)

The subject, site personnel, and the Sponsor were blinded to the subject's study drug assignment. Study monitors, data management, and biostatistics personnel were unaware of the contents of the randomization code until all data were entered and validated and the database was locked.

All subjects were treated with istradefylline 20 mg/d, 40 mg/d, or matching placebo tablets for 12 weeks. As the istradefylline 20 and 40 mg tablets were different in size, all subjects also received a placebo tablet for the alternate dosage; subjects randomized to placebo received two placebo tablets

Statistical methods

There were a large number of changes to the statistical analysis. The original SAP for this study was developed and finalised on 02 Apr 2014. Four SAP amendments were created and signed on 22 Oct 2014 (Amendment 1), 11 Dec 2015 (Amendment 2), 13 Jul 2016 (Amendment 3), and 11 Nov 2016 (Amendment 4). Database lock occurred on 14 Nov 2016. The final SAP (6002-014-statistical-analysis-plan-amendment04-en) supersedes the statistical considerations identified in the clinical protocol.

The following analysis populations were planned to be used in the study: safety analysis set and ITT set as specified for other pivotal trials and an efficacy evaluable set including all subjects completing 12 weeks of treatment with no major protocol violations. This population will be specified before the study database is locked and unblinded. Major protocol violations are identified by medical review of the study

protocol deviation log maintained by clinical operations. The categories of major protocol violations include: change to anti-parkinsonian medication; invalid baseline diary data; inclusion/exclusion criteria non-compliance and study procedure non-compliance.

All efficacy data collected in this study will be summarized using descriptive statistics at each assessment time for each treatment group based on actual values and change from Baseline values. Continuous variables will be summarized using n, mean, SD, median, minimum, and maximum values. Categorical variables will be summarized using the number and percentage of subjects in each category.

Subjects will complete three 24-hour ON/OFF patient diaries on the 3 consecutive days immediately prior to each scheduled visit including Baseline. The overall diary response for that visit will be the average of the values within each diary category over all completed valid diaries at the visit. Note that a valid diary is defined as a completed diary with no more than 4 invalid time points as defined below. The total hours in any particular state for a visit will be the average across all valid diaries collected that week, thus averaging over either 1, 2, or 3 valid diaries. If all three completed diaries within the same week are invalid, then the overall diary entry for that week will be considered missing. The total hours and percentage of awake time per day spent in the OFF state for the visit will be calculated as an average of the values over all valid diaries completed at the visit. The change from Baseline in total hours and change from Baseline in percentage of awake time per day spent in the OFF state will then be calculated.

Two approaches will be used for handling invalid entries on 24 hour diaries. An observed case analysis (primary) will treat all invalid entries (i.e., missing evaluation or more than one entry) as missing. A worst-case analysis will impute OFF for all invalid entries within experimental groups and impute ON without dyskinesia for all invalid entries within the placebo group.

The primary efficacy variable was planned to be analysed using a MMRM approach with Baseline value as a covariate, fixed effect terms in the model for study site, treatment group, week, and treatment-by-week interaction, and an unstructured covariance matrix. Using this model, the difference in change from Baseline to Week 12 in total hours OFF time between the 40 mg/d treatment group and the placebo treatment group was to be tested using a t-test at the two-sided 5% significance level. In the event that the unstructured covariance matrix is non-estimable (due to lack of convergence) when fitting the MMRM to the observed study data, an AR(1) covariance matrix will be used instead.

A simple hierarchical sequence of statistical testing was to be used for the primary efficacy endpoint and the key secondary efficacy endpoint for both doses using a two-sided test at a critical alpha of 0.05 as follows:

Step 1. Change from Baseline to Week 12 in Total "OFF" time for 40 mg/d versus placebo.

Step 2. Change from Baseline to Week 12 in Total "OFF" time for 20 mg/d versus placebo.

Step 3. Change from Baseline to Week 12 in Total "ON" time without troublesome dyskinesia for 40 mg/d versus placebo.

Step 4. Change from Baseline to Week 12 in Total "ON" time without troublesome dyskinesia for 20 mg/d versus placebo.

Comparison of change from baseline for this key secondary endpoint between the 40 mg/d and placebo treatment arms and between the 20 mg/d and placebo treatment arms were evaluated in the ITT group using a t-test from the MMRM.

Several secondary analyses were conducted to address regional or country effects on the primary endpoint. The primary endpoint was also analysed using a MMRM approach with baseline value as a covariate, fixed effect terms in the model for country (United States, Canada, Czech Republic, Germany, Israel, Italy, Poland, and Serbia), treatment group, week, and treatment-by-week interaction, and an

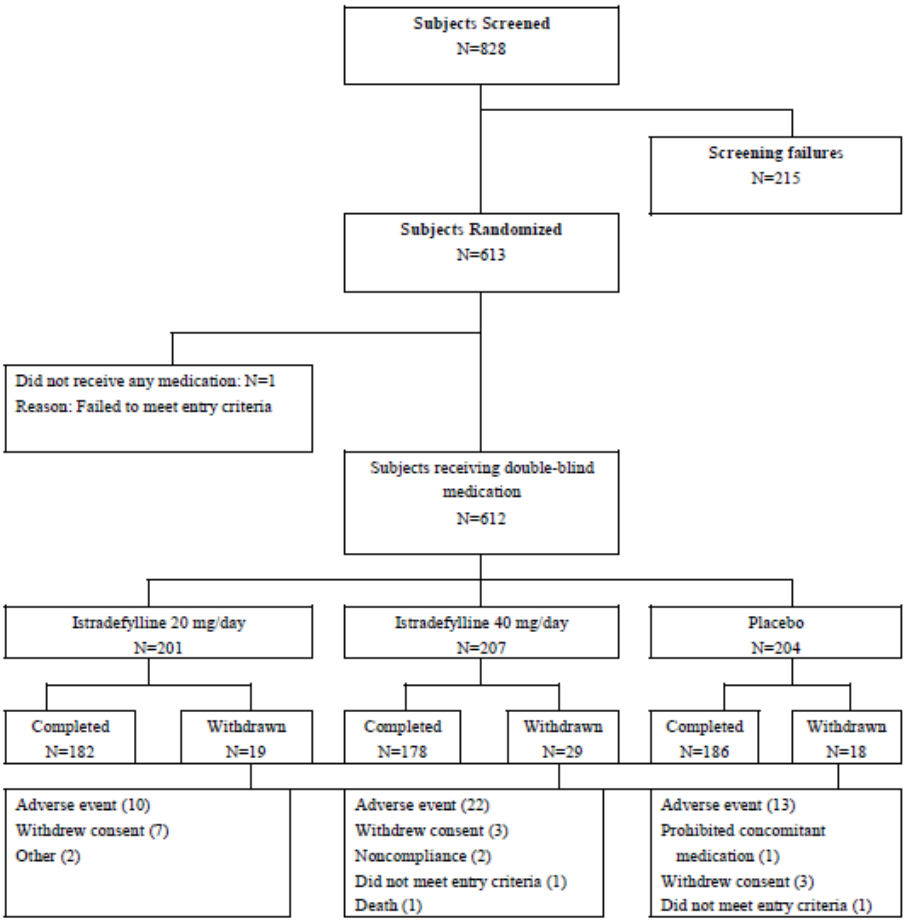
unstructured covariance matrix. A separate analysis was run using the same model mentioned above, but replacing the fixed effect term of country with region (North America [United States and Canada] or Rest of World [remaining countries]). In each analysis, the term for country or region was tested for significance in the model.

A pooling strategy for study sites was to be implemented at the end of the study for any site with less than 12 subjects randomized (fewer than 2% of all randomized subjects). Sites are pooled based on geographic region or type of investigational site (e.g., academic or private practice). The size of any pooled set of sites would not be larger than the total number of randomized subjects at the largest freestanding site. The pooling strategy is documented prior to unblinding of the treatment codes. This collection of pooled sites was to be used as the fixed effect term "Site" in the MMRM.

Results

Participant flow

Figure 42: Disposition of subjects



Recruitment

Study locations: 47 sites in the US, 6 in Czech Republic, 13 in Germany, 9 in Italy, 7 in Poland, 4 in Serbia, 5 in Canada and 5 in Israel

Study period: first patient entered the study on 5 November 2013 and last patient out 23 September 2016

Conduct of the study

The protocol was amended 4 times. Amendments #1, #2 and#3 were approved prior to the entry of the first patient to the study and will not be further discussed. Amendment #4 was approved in the month before the last patient completed. Amendment #4, 15 August 2016 (last subject completed (23 September 2016)

There were 4 amendments to the statistical analysis plan.

Changes to the statistical wording of the protocol following changes to the SAP following dialogue with the FDA

- Total hours of ON time/day without troublesome dyskinesia was designated as the key secondary objective or endpoint throughout the protocol.
- The justification of the sample size text was updated regarding the primary efficacy variable per the FDA request to change one-sided 2.5% significance level to two-sided 5% significance level. In

addition, "approximately" was added in the following: "The total sample size used for this study will then be approximately 609 subjects, 203 subjects per treatment group."

- The Analysis "Populations" were changed to Analysis "Sets". The definitions of the ITT and EE populations were updated:

  a) from: ITT including all subjects randomized to a treatment arm and assigned a study number to ITT including all subjects randomized to a treatment arm and with both a valid baseline and at least one valid post-baseline patient diary.

  b) from: EE set including all subjects completing 12 weeks of treatment with no major protocol violations. If applicable, this population will be specified before the study database is locked and unblinded to EE set including all subjects completing 12 weeks of treatment with no major protocol violations. This population will be specified before the study database is locked and unblinded. Major protocol violations are identified by medical review of the study protocol deviation log maintained by clinical operations.

- The information relative to the presentation of subject disposition was updated.
- The information relative to the presentation of demographic and baseline characteristics was updated to be summarized and based on the ITT, EE, and safety analysis sets.
- The information relative to the presentation of concomitant medications was updated.
- The information relative to the presentation of study drug exposure and compliance was updated to be summarized by treatment group as the number of weeks (defined as [days from first dosing to last dosing]/7) receiving randomized treatment from the first day of dosing until the last day of dosing.
- Text regarding diary completion and the definition of their validity was updated.
- A sensitivity analysis of the primary efficacy variable was specified as "The primary efficacy analysis will be based on the ITT analysis set. A sensitivity analysis of the primary efficacy variable will also be conducted based on the observed case analysis with the EE analysis set.
- The calculation of UPDRS subscale scores was updated to include total score (for both Parts I, II, III, and total). ESS was updated to ESS total score.
- The step-down approach to control the experiment-wise error rate was deleted and replaced with a simple hierarchical sequence of statistical testing.
- The descriptions of secondary efficacy analyses, subgroup analyses using subject characteristic variables, updates to the pooling strategy, and sensitivity analyses were rewritten.
- The definition of TEAEs was updated to those AE with an onset time on or after the start of study drug, or are ongoing at the time of study drug initiation and increase in severity or become closer in relationship to study drug during the treatment period.
- Safety parameters were clarified relative to missing start dates to use the worst-case approach, clarification for the counting of TEAEs, the display of TEAEs, the analysis of C-SSRS data, the definition of the definition of out of normal range laboratory values and their display and analysis.
- A statement was added to note that the details of the PK analyses will be described in a separate PK/PD analysis plan.

Baseline data

Baseline demographic features were similar across treatment groups. PD characteristics were also broadly similar across treatment groups e.g. time since diagnosis, duration of levodopa therapy, dose of levodopa, duration of motor complications, H&Y and UPDRS 3 scores. In terms of mean duration of OFF time this was slightly greater in the placebo arm than in either istradefylline arm.

Table 87: Demographic and disease characteristics at baseline

	Placebo N = 204	Istradefylline 20mg/day N = 201	Istradefylline 40mg/day N = 207
Age Mean (SD) Median (Min, Max)	 63.8(8.49) 64 (41, 86)	 63.5 (8.65) 64 (41, 87)	 64.5 (8.17) 64 (40, 87)
Male Female	124 (60.8%) 80 (39.2%)	125 (62.2%) 76 (37.8%)	126 (60.9%) 81 (39.1%)
Time since diagnosis Years 1-3 4-7 ≥ 8	 7 (3.4%) 66 (32.4%) 131 (64.2%)	 10 (5%) 76 (37.8%) 115 (57.2%)	 9 (4.3%) 66 (31.9%) 132 (63.8%)
Duration levodopa (years) Mean (SD) Median (Min, Max)	N = 204 8.9 (4.03) 8 (2, 23)	N = 201 8.5 (4.58) 7 (2, 24)	N = 206 8.9 (4.58) 8 (2, 28)
Mean daily dose LD (mg) Mean (SD) Median (Min, Max)	 815.02 (385.021) 712.5 (400, 3150)	 834.89 (360.769) 775 (400, 2500)	 841.81 (394.431) 800 (300, 3150)
Duration motor complications Mean (SD) Median (Min, Max)	N = 203 6.1 (4.32) 5 (1, 25)	N = 200 5.7 (3.98) 4.5 (1, 21)	N = 204 6.3 (4.39) 5 (1, 22)
Total hours awake Off Mean (SD) Median (Min, Max)	 5.41 (2.015) 5.17 (1.8, 14.3)	 5.37 (1.975) 5.33 (1, 12.2)	 5.19 (2.077) 4.83 (1, 11)
Total hours ON without troublesome dyskinesia Mean (SD) Median (Min, Max)	 9.25 (2.586) 9.42 (1, 14.8)	 9.72 (2.305) 9.83 (1.8, 15)	 9.65 (2.383) 9.67 (1.2, 16)
UODRS Part 111 Mean (SD) Median (Min, Max)	N = 204 21.8 (10.76) 20.5 (3, 68)	N = 201 22.7 (11.52) 22 (4, 62)	N = 206 23.4 (12.03) 22 (2, 64)
Hoehn & Yahr ON Stage 2 Stage 2.5 Stage 3 Stage 4	 91 (44.6%) 50 (24.55) 56 (27.5%) 7 (3.4%)	 94 (46.8%) 55 (37.4%) 49 (24.4%) 3 (1.5%)	 96 (46.4%) 54 (26.1%) 47 (22.7%) 10 (4.8%)

28.1% overall H&Y stage 3 or 4.

Forty-five percent of the randomised population were recruited in the US (n = 278), 44% in Europe (n = 267) and the remainder in Israel and Canada. Recruitment by country is shown in Table 88.

Table 88: Randomised population by country

	Placebo	Istradefylline 20mg	Istradefylline 40mg	Total
US	93	93	92	278
Czech Republic	22	23	23	68
Germany	13	15	17	45
Italy	21	18	20	59
Poland	24	23	23	70
Serbia	9	7	9	25
Israel	11	12	12	35
Canada	11	11	11	33
Total	204	202	207	613

Of note at least one third of patients in each treatment group were also receiving entacapone with a somewhat higher rate in those in the istradefylline 40mg group (32.8% P, 35.3% Istra 20mg, 39.1% Istra 40mg)

This population appears to be a more severely affected population than that of EU-007 and is more heavily treated with adjunctive therapies.

Table 89: Prior antiparkinsonian medications taken by ≥ 5% of subjects in any treatment group (safety analysis set)

Therapeutic Class[a]	Placebo (N=204)	Istradefylline 20 mg/day (N=201)	Istradefylline 40 mg/day (N=207)	Total (N=612)
Took any antiparkinsonian medication	204 (100.0)	201 (100.0)	207 (100.0)	612 (100.0)
Levodopa	204 (100.0)	201 (100.0)	207 (100.0)	612 (100.0)
Carbidopa	169 (82.8)	172 (85.6)	184 (88.9)	525 (85.8)
Entacapone	67 (32.8)	71 (35.3)	81 (39.1)	219 (35.8)
Ropinirole hydrochloride	56 (27.5)	47 (23.4)	61 (29.5)	164 (26.8)
Pramipexole dihydrochloride	50 (24.5)	58 (28.9)	44 (21.3)	152 (24.8)
Benserazide	41 (20.1)	46 (22.9)	42 (20.3)	129 (21.1)
Rasagiline mesylate	44 (21.6)	36 (17.9)	46 (22.2)	126 (20.6)
Amantadine hydrochloride	39 (19.1)	25 (12.4)	42 (20.3)	106 (17.3)
Ropinirole	23 (11.3)	28 (13.9)	22 (10.6)	73 (11.9)
Rotigotine	20 (9.8)	20 (10.0)	22 (10.6)	62 (10.1)
Amantadine	19 (9.3)	22 (10.9)	17 (8.2)	58 (9.5)
Rasagiline	17 (8.3)	18 (9.0)	16 (7.7)	51 (8.3)
Selegiline hydrochloride	17 (8.3)	15 (7.5)	11 (5.3)	43 (7.0)
Amantadine sulfate	12 (5.9)	14 (7.0)	14 (6.8)	40 (6.5)
Pramipexole	7 (3.4)	10 (5.0)	11 (5.3)	28 (4.6)

Note: Includes medications taken within 30 days prior to the first dose of double-blind study drug.
a: WHODRUG Dictionary Version 2013 SEP was used for coding.

Numbers analysed

All efficacy analyses derived from the 24-hour ON/OFF patient diary were carried out in the ITT population. The primary endpoint (change from baseline in the total hours of awake time/day spent in the OFF state) and the key secondary endpoint (change from baseline in total hours of ON time without troublesome dyskinesia) were analysed in the ITT (all those randomised with a valid baseline and at

least 1 valid post-baseline diary) population. Sensitivity analyses of the primary endpoint and the key secondary endpoint were also performed in the ITT population using the worst case and placebo multiple imputation approaches.

Table 90: Patient disposition and analysis populations by treatment group

	Placebo	Istradefylline 20 mg	Istradefylline 20 mg
Randomised	204	202	207
Safety[a]	204	201	207
ITT[b]	198 (97.1%)	194 (96%)	200 (96.6%)
EE[c]	172 (84.3%)	165 (81.7%)	159 (76.8%)
Completed study	186 (91.2%)	182 (90.1%)	178 (86%)

a: Includes all subjects who received at least one dose of assigned study drug (even a partial dose).
b: Includes all subjects randomized and had both a valid baseline and at least one valid post-baseline patient diary.
c: Includes all subjects all subjects from the Intent-to-treat analysis set completing 12 weeks of treatment with no major protocol violations.

Outcomes and estimation

Primary efficacy endpoint

The primary efficacy endpoint was the change from baseline in the total hours of awake time/day spent in the OFF state at Week 12 based on the 24-hour ON/OFF patient diary data. As can be seen from the following Table the primary endpoint was not achieved for Istradefylline 40mg/day nor for Istradefylline 20mg/day. LSM change from baseline for placebo at week 12 was -0.88 hours, for Istradefylline 20mg/day -1.2 hours and for Istradefylline 40mg/day -1.15 hours. The difference in LSM change from baseline to placebo was -0.32 hours (95% CI -0.76, 0.12) for istradefylline 20mg and -0.27 hours (95% CI -0.7, 0.17) for istradefylline 40mg/day (See following

Table 91).

Note the percentage with 3 valid diaries at week 12 was highest in the placebo group (93.9%) and lowest in the istradefylline 40mg/day group (86%).

Table 91: Total Hours of Awake Time/day Spent in the OFF State Based on Patient's ON/OFF Diary - Actual and Change from Baseline (observed Case Analysis)

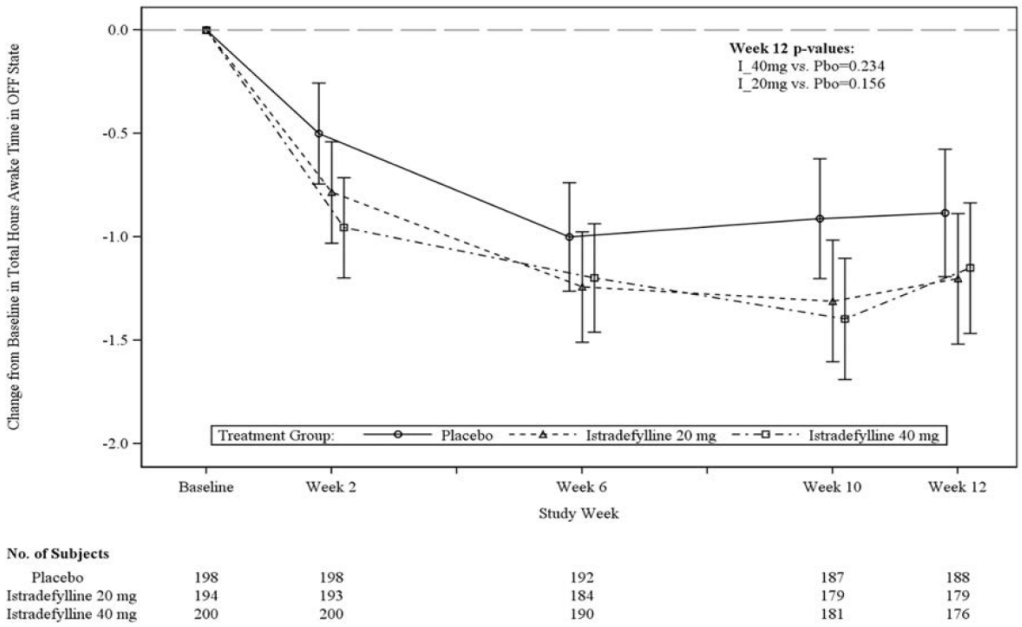
	Placebo N = 198		Istradefylline 20mg/day N = 194		Istradefylline 40mg/day N = 200	
Baseline	Actual	Change	Actual	Change	Actual	Change
Mean (SD)	5.44 (2.005)		5.4 (1.958)		5.23 (2.059)	
Median (Min, Max)	5.17 (2, 14.3)		5.33 (1, 12.2)		4.92 (1.2, 11)	
Week 12						
No. with 3 valid diaries	186 (93.9%)	186	173 (89.2%)	173	172 (86%)	172
Mean (SD)	4.52 (2.832)	-0.91	4.13 (2.372)	-1.26 (2.405)	4.16 (2.542)	-1.13 (2.061)
Median (Min. Max)	4.17 (0, 16.7)	-0.92 (-8.5, 5.2)	4 (0, 11.3)	-1 (-9.3,5.3)	3.83 (0, 13.8)	-1.25 (-6.7, 6)
LSM 95% CI of change	4.48	-0.88 -1.19, -1.58	4.17	-1.2 -1.52, -0.89	4.22	-1.15 -1.46, -0.84

Diff LSM[a] (95% CI)				-0.32 (-0.76, 0.12)		-0.27 (-0.7, 0.17)
P values				0.156		0.234

Note: Invalid entries on a diary were treated as missing.
a: Difference in LSM, corresponding 95% CI, and p-values are from paired comparisons between each istradefylline treatment arm vs placebo. A mixed model repeated measures approach was used with baseline assessment as a covariate, and pooled study site, treatment group, week, and treatment-by-Week interaction as fixed effect terms.
CI=confidence interval; LS=least squares; max=maximum; min=minimum; SD=standard deviation

Whilst the change from baseline is numerically greater at all non-baseline time points for both istradefylline groups than placebo however these differences vary by time point and appear to be greatest at week 2, lowest at week 6 and at endpoint.

Figure 43: primary endpoint: change from baseline (LSM with 95%CI) in total hours/day spent in the OFF state (observed case analysis)



Secondary efficacy endpoints

This study failed to demonstrate any efficacy for istradefylline 40mg over placebo in the primary endpoint. Therefore, results for secondary efficacy endpoints are not further considered (percentage of awake time / day spent ON without troublesome dyskinesia is provided in the table below for completeness as a relevant secondary efficacy endpoint).

Ancillary analyses

A number of sub-group analyses were carried out, given the failure of the study to meet the primary endpoint and the hierarchical approach to statistical testing, these are not further discussed.

Summary of main efficacy results

This summary should be read in conjunction with the discussion on clinical efficacy as well as the benefit risk assessment (see later sections).

Table 92: Summary of efficacy for trial 6002-014

Title: A Phase 3, 12-week, Double-blind, Placebo-controlled, Randomized, Multicenter Study to Evaluate the Efficacy of Oral Istradefylline 20 and 40 mg/day as Treatment for Subjects with Moderate to Severe Parkinson's Disease					
Study identifier	6002-014				
Design	Phase 3, randomised, double-blind, placebo-controlled, fixed-dose, parallel-group, multicenter trial.				
	Duration of main phase:		12 weeks		
	Duration of Run-in phase: Duration of Extension phase:		Not applicable Not applicable		
Hypothesis	Superiority				
Treatments groups	Istradefylline 20 mg		Istradefylline 20 mg once daily, 12 weeks, n=202 subjects		
	Istradefylline 40 mg		Istradefylline 40 mg once daily, 12 weeks, n=207 subjects		
	Placebo		Placebo once daily, 12 weeks, n=204 subjects		
Endpoints and definitions	Primary endpoint	OFF time per day	Change from baseline in total hours of awake time/day spent in the OFF state		
	Key secondary endpoint	ON time per day without troublesome dyskinesia	Change from baseline in total hours of ON time/day without troublesome dyskinesia		
Database lock	14 November 2016				
Results and Analysis					
Analysis description	**Primary Analysis**				
Analysis population and time point description	Intent to Treat (ITT), week 12 Observed case, MMRM				
Descriptive statistics and estimate variability	Treatment group		Placebo	Istradefylline 20 mg	Istradefylline 40 mg
	Number of subjects analysed at Week 12		188	179	176
	Change from baseline in total hours of awake time/day spent in the OFF state; Mean (SD)		-0.91 (2.186)	-1.26 (2.405)	-1.13 (2.061)
	Range (min to max)		-8.5 to 5.2	-9.31 to 5.3	-6.7 to 6.0
	Change from baseline in total hours of ON time/day without troublesome dyskinesia; Mean (SD)		0.90 (2.529)	1.01 (2.697)	0.79 (2.536)
	Range (min to max)		-6.7 to 9.2	-8.7 to 9.2	-11.5 to 7.7
Effect estimate per comparison	Primary endpoint (hours/day spent in the OFF state); difference from		Comparison groups	Istradefylline 20 mg vs Placebo	Istradefylline 40 mg vs Placebo

	placebo in the LSM change from baseline	LSM change vs. placebo	-0.32	-0.27
		95% CI	-0.76, 0.12	-0.70, 0.17
		P-value	0.156	0.234
	Key secondary endpoint (hours/day spent ON without troublesome dyskinesia); difference from placebo in the LSM change from baseline	Comparison groups	Istradefylline 20 mg vs Placebo	Istradefylline 40 mg vs Placebo
		LSM change vs. placebo	0.24	0.00
		95% CI	-0.28, 0.77	-0.52, 0.53

Clinical studies in special populations

No additional efficacy studies were performed in special populations

Analysis performed across trials (pooled analyses and meta-analysis)

Pooled results help to illustrate the redistribution of the 24-hour day among OFF time and categories of ON time with istradefylline treatment, relative to placebo. For the pool of 8 RCTs, istradefylline decreased time spent in the OFF state with both 20 and 40 mg/day. With no notable change in daily time asleep (row B), the istradefylline-associated decreases in OFF time per day were largely converted to ON time without troublesome dyskinesia ("good ON", row H). The small increases in ON with troublesome dyskinesia (row F, corresponding to 5 to 8 minutes/day) were overshadowed by the increases in ON without troublesome dyskinesia (row H). Relative to placebo, istradefylline had little effect on UPDRS Part II ON (row I). Nevertheless, there were modest, decreases (improvement) in UPDRS Part III ON (row J).

Table 93: Pooled results for 8 RCTs-Difference from placebo in the change from baseline to week 12 for diary-related and UPDRS Endpoints (OC, ITT, MMRM)

		LS Mean Difference from Placebo hours (95% CI)	
	Efficacy Endpoint	**Istradefylline 20 mg/day**	**Istradefylline 40 mg/day**
A	Total OFF	-0.38 (-0.61, -0.15)	-0.45 (-0.68, -0.22)
B	Asleep time	-0.11 (-0.23, 0.01)	-0.02 (-0.14, 0.10)
C	Total ON	0.48 (0.24, 0.71)	0.45 (0.22, 0.69)
D	ON without dyskinesia	0.15 (-0.13, 0.44)	0.07 (-0.22, 0.35)
E	ON with dyskinesia	0.34 (0.09, 0.58)	0.39 (0.15, 0.63)
F	ON with troublesome dyskinesia	0.08 (-0.06, 0.21)	0.13 (-0.00, 0.26)
G	ON with non-troublesome dyskinesia	0.25 (0.04, 0.45)	0.25 (0.05, 0.45)
H	ON without troublesome dyskinesia	0.40 (0.15, 0.66)	0.33 (0.08, 0.59)
I	UPDRS Part II (score)	0.1 (-0.2, 0.4)	-0.2 (-0.5, 0.1)
J	UPDRS Part III (score)	-0.6 (-1.3, 0.1)	-1.3 (-2.0, -0.6)

CI= Confidence interval; ITT= Intention-to-treat; LS=Least squares; MMRM=Mixed-model repeated measures; OC= Observed case; RCT=randomized controlled trial; UPDRS=Unified Parkinson Disease Rating Scale.

Age Subgroup Analyses for 8 Study Pool

Table 94: Total hours per day in the OFF State at Week 12: age subgroups (<65 and ≥65 years) Actual and Change from baseline (observed case analysis in the ITT set).

Age Group: < 65 Years

	Placebo (N=530)		Istradefylline 20 mg/day (N=417)		Istradefylline 40 mg/day (N=464)	
Visit Statistics	Actual	Change	Actual	Change	Actual	Change
Week 12						
n	485	485	382	382	417	417
Mean	5.35	-0.89	5.05	-1.29	5.24	-0.97
SD	2.881	2.368	2.696	2.412	2.865	2.432
Median	5.30	-0.70	5.00	-1.13	5.30	-0.83
Min	0.0	-10.8	0.0	-12.5	0.0	-8.8
Max	16.7	8.0	14.6	7.2	15.3	6.8
LS Mean	5.43	-0.82	5.12	-1.14	5.26	-0.99
95% CI (of change)		(-1.04,-0.61)		(-1.39,-0.88)		(-1.23,-0.75)
Diff in LS Means [a]				-0.31		-0.17
95% CI [a]				(-0.63,0.01)		(-0.48,0.15)

Age Group: >= 65 Years

	Placebo (N=462)		Istradefylline 20 mg/day (N=431)		Istradefylline 40 mg/day (N=415)	
Visit Statistics	Actual	Change	Actual	Change	Actual	Change
Week 12						
n	417	417	392	392	381	381
Mean	5.39	-0.83	4.76	-1.33	4.59	-1.45
SD	3.100	2.691	2.947	2.639	2.654	2.489
Median	5.20	-0.80	4.37	-1.20	4.50	-1.50
Min	0.0	-14.0	0.0	-13.3	0.0	-10.7
Max	14.8	8.3	16.8	9.8	13.8	6.5
LS Mean	5.43	-0.70	4.88	-1.25	4.51	-1.62
95% CI (of change)		(-0.95,-0.46)		(-1.51,-0.99)		(-1.89,-1.36)
Diff in LS Means [a]				-0.55		-0.92
95% CI [a]				(-0.90,-0.20)		(-1.27,-0.57)

Table 95: Total hours per day in the OFF State at Week 12: age subgroups (<75 and ≥75 years) Actual and Change from baseline (observed case analysis in the ITT set).

Age Group: < 75 Years

	Placebo (N=882)		Istradefylline 20 mg/day (N=735)		Istradefylline 40 mg/day (N=790)	
Visit Statistics	Actual	Change	Actual	Change	Actual	Change
Week 12						
n	802	802	671	671	717	717
Mean	5.34	-0.88	4.84	-1.38	4.98	-1.18
SD	2.917	2.502	2.770	2.462	2.813	2.480
Median	5.17	-0.70	4.80	-1.20	5.00	-1.17
Min	0.0	-14.0	0.0	-13.3	0.0	-10.7
Max	16.7	8.3	14.6	7.2	15.3	6.8
LS Mean	5.33	-0.87	4.92	-1.28	4.94	-1.26
95% CI (of change)		(-1.03,-0.70)		(-1.47,-1.10)		(-1.44,-1.08)
Diff in LS Means [a]				-0.42		-0.39
95% CI [a]				(-0.66,-0.17)		(-0.63,-0.15)

Age Group: >= 75 Years

Visit Statistics	Placebo (N=110) Actual	Placebo (N=110) Change	Istradefylline 20 mg/day (N=113) Actual	Istradefylline 20 mg/day (N=113) Change	Istradefylline 40 mg/day (N=89) Actua l	Istradefylline 40 mg/day (N=89) Change
Week 12						
n	100	100	103	103	81	81
Mean	5.61	-0.73	5.26	-0.87	4.50	-1.31
SD	3.467	2.676	3.170	2.895	2.485	2.385
Median	5.55	-0.80	4.80	-0.50	4.50	-1.40
Min	0.0	-9.5	0.0	-12.0	0.0	-8.3
Max	14.6	5.5	16.8	9.8	9.5	5.1
LS Mean	5.52	-0.60	5.37	-0.76	4.82	-1.31
95% CI (of change)		(-1.21,-0.00)		(-1.37,-0.14)		(-2.00,-0.61)
Diff in LS Means *				-0.15		-0.71
95% CI *				(-1.02,0.71)		(-1.67,0.26)

Race Subgroup Analysis 8 Study Pool

Table 96: Total hours per day in the OFF State at Week 12: race subgroups Actual and Change from baseline (observed case analysis in the ITT set).

Race: Caucasian

Visit Statistics	Placebo (N=668) Actual	Placebo (N=668) Change	Istradefylline 20 mg/day (N=584) Actual	Istradefylline 20 mg/day (N=584) Change	Istradefylline 40 mg/day (N=560) Actual	Istradefylline 40 mg/day (N=560) Change
Week 12						
n	607	607	534	534	506	506
Mean	5.20	-1.00	4.75	-1.34	4.81	-1.24
SD	2.951	2.608	2.724	2.673	2.802	2.517
Median	5.00	-0.83	4.50	-1.17	4.58	-1.20
Min	0.0	-14.0	0.0	-13.3	0.0	-8.8
Max	16.7	8.3	16.8	9.8	15.3	6.8
LS Mean	5.21	-0.90	4.93	-1.18	4.84	-1.27
95% CI (of change)		(-1.10,-0.70)		(-1.41,-0.96)		(-1.50,-1.05)
Diff in LS Means *				-0.28		-0.37
95% CI *				(-0.57,0.01)		(-0.67,-0.08)

Race: Non-Caucasian

Visit Statistics	Placebo (N=324) Actual	Placebo (N=324) Change	Istradefylline 20 mg/day (N=264) Actual	Istradefylline 20 mg/day (N=264) Change	Istradefylline 40 mg/day (N=319) Actual	Istradefylline 40 mg/day (N=319) Change
Week 12						
n	295	295	240	240	292	292
Mean	5.72	-0.59	5.23	-1.24	5.14	-1.11
SD	3.022	2.312	3.025	2.174	2.743	2.386
Median	5.40	-0.50	5.15	-1.20	5.35	-0.95
Min	0.0	-8.2	0.0	-11.0	0.0	-10.7
Max	15.8	6.0	14.6	6.8	14.4	5.3
LS Mean	5.51	-0.84	4.87	-1.48	4.89	-1.47
95% CI (of change)		(-1.18,-0.50)		(-1.86,-1.11)		(-1.82,-1.11)
Diff in LS Means *				-0.64		-0.63
95% CI *				(-1.02,-0.26)		(-0.98,-0.27)

Gender Subgroup analysis for 8 study pool

Table 97: Total hours per day in the OFF State at Week 12: gender subgroups Actual and Change from baseline (observed case analysis in the ITT set).

Gender: Female

	Placebo (N=413)		Istradefylline 20 mg/day (N=362)		Istradefylline 40 mg/day (N=367)	
Visit Statistics	Actual	Change	Actual	Change	Actual	Change
Week 12						
n	377	377	330	330	328	328
Mean	5.57	-0.58	4.90	-1.39	4.78	-1.35
SD	2.984	2.388	2.811	2.377	2.749	2.472
Median	5.30	-0.50	4.80	-1.30	4.83	-1.20
Min	0.0	-9.0	0.0	-13.3	0.0	-10.7
Max	16.7	8.3	14.6	7.2	15.3	6.3
LS Mean	5.63	-0.56	4.96	-1.23	4.86	-1.34
95% CI (of change)		(-0.82,-0.31)		(-1.51,-0.96)		(-1.61,-1.06)
Diff in LS Means *				-0.67		-0.77
95% CI *				(-1.03,-0.31)		(-1.13,-0.42)

Gender: Male

	Placebo (N=579)		Istradefylline 20 mg/day (N=486)		Istradefylline 40 mg/day (N=512)	
Visit Statistics	Actual	Change	Actual	Change	Actual	Change
Week 12						
n	525	525	444	444	470	470
Mean	5.23	-1.06	4.90	-1.25	5.03	-1.09
SD	2.976	2.596	2.843	2.635	2.806	2.465
Median	5.00	-1.00	4.63	-1.00	5.00	-1.17
Min	0.0	-14.0	0.0	-12.5	0.0	-9.1
Max	16.0	8.0	16.8	9.8	15.0	6.8
LS Mean	5.21	-0.98	5.03	-1.17	4.95	-1.24
95% CI (of change)		(-1.20,-0.77)		(-1.41,-0.92)		(-1.47,-1.00)
Diff in LS Means *				-0.18		-0.25
95% CI *				(-0.50,0.13)		(-0.57,0.06)

2.5.2. Discussion on clinical efficacy

Design and conduct of clinical studies

In support of the application the applicant submitted 4 studies which could be categorised as dose finding studies, 8 pivotal studies, 3 follow up studies and further pooled analyses of the results of all of the pivotal studies and additionally of the 5 studies in which they considered efficacy was shown.

Dose finding studies

The CHMP considered the following four studies as Dose-Finding Studies: 6002-EU04; 6002-EU05; 6002-US-001; 6002-US004. In addition, there was an objective of dose finding in the pivotal studies; 6002-US-018, 6002-0608 and 6002-EU-007.

Study 6002-US-001, is considered most relevant for efficacy assessment, in that this 12-week DB, placebo controlled, randomised parallel group multi centre exploratory safety and efficacy study of KW-6002 enrolled 83 patients with somewhat similar inclusion criteria to the subsequent pivotal studies and

KW-6002 was administered as adjunctive therapy to L/C. The study involved 3 treatment periods of 4 weeks duration each and explored three dose groups of 5/10/20mg, 10/20/40mg and placebo throughout. Relevant to efficacy assessment in the pivotal trials, the % change from baseline in proportion of awake time spent in an OFF state based on investigator's ON/OFF assessment as recorded in an 8 hour evaluation showed significant difference; comparisons of KW-6002 with placebo at endpoint had p values of 0.004 (combined) and 0.007 (5/10/20 mg/day) and 0.024 (10/20/40 mg/day) for each dose groups respectively. Of note there was a slightly greater reduction in time spent in the OFF state for the lower dose sequence 5/10/20 compared to higher dose 10/20/40mg, however due to the limited sample size this may have been attributable to chance and may highlight the limited potential of these earlier studies to conclusively determine the presence or absence of a dose response relationship.

Overall, the rationale in progression of conducting the 'dose-finding' studies is not entirely clear. Different dose schedules including different titration periods are noted in each of the four studies.

No conclusions were determined from these earlier studies and pivotal trials went on to repeatedly explore doses of 10mg, 20mg, 40mg and 60mg, in a rather unclear sequence.

The applicant has not provided conclusive evidence to support the recommendation to increase the Istradefylline dose from 20 to 40 mg/day, considering the observed results in the pivotal studies are inconsistent in showing any increased efficacy of the 20mg dose compared to 40mg dose.

Pivotal studies

All eight pivotal studies were randomised placebo-controlled trials. Patients continued with their stable anti-PD therapy over the duration of the study. Study 6002-EU-007 was the only study that had an active comparator arm (entacapone) in addition to the placebo arm, which fulfilled the requirement in the Guideline on clinical investigation of medicinal products in the treatment of Parkinson's disease EMA/CHMP/330418/2012 rev. 2 to include a three arm study design.

The applicant sought scientific advice from CHMP in 2004. In general, the applicant adhered to the advice. However, the CHMP had recommended performing the study in patients receiving L-dopa only, yet all 8 pivotal trials also allowed additional concomitant dopaminergic medications.

Four of the studies (Studies 6002-US-005; 6002-US-006; 6002-US-013; 6002-US-018) were conducted in North America between 2002 and 2005. One study (Study 6002-EU-007) was conducted at a number of sites in Europe, as well as Russia, India, South Africa, Chile and Argentina between 2004 - 2005. Two studies were performed in Japan in 2007-08 (Study 6002-0608) and 2009-11 (Study 6002-009). The final study was performed in the US, Canada, the EU, Serbia and Israel over the course of 2013 to 2016 (Study 6002-014).

It is questionable how generalisable the results of these studies are to today's PD population given the wider range of adjunctive therapy available today and changing practices in the management of PD. In the US studies, 40-50% of patients received concomitant COMT inhibitors, whereas in EU-007 no patients on the istradefylline arm received COMT inhibitors, as Entacapone was the active comparator.

In the Japanese studies Study 6002-0608 and Study 6002-009 there were a higher proportion of patients who received dopamine agonists, MAO-B inhibitors, amantadine derivatives and anticholinergics compared to the other 6 studies.

There were a number of commonalities across studies regarding inclusion/exclusion criteria. All participants were required to: meet the UKPDS Brain Bank diagnostic criteria (step 1 and 2) for PD; have predictable wearing OFF periods varying from an average of 2 or 3 hours per days recorded in patient diaries prior to randomization; have a modified H&Y score while in the OFF state of II to IV (all except Study 6002-014); be responsive to levodopa with a decarboxylase inhibitor, with at least a 1 year history of treatment with levodopa and on a stable regimen for at least 4 weeks at baseline. The minimum

number of daily levodopa doses was three in all of the pivotal studies, apart from Study 6002-014, where it was 4.

In studies 6002-0608 and 6002-009, a minimum daily dose of 300mg levodopa was required and in study 014 a minimum dose of 400mg levodopa was required. No minimum dose was specified in the other 5 pivotal studies. For all pivotal studies participants were required to meet diary training requirements. In PD clinical trials investigating the efficacy of medicinal products in reducing OFF periods, it is standard practice to collect data on the duration of OFF and ON periods with and without dyskinesia in a patient completed diary. This is in line with the recommendations of the *Guideline on clinical investigation of medicinal products in the treatment of Parkinson's disease EMA/CHMP/330418/2012 rev. 2*. The Hauser diary was used in all of the studies.

OFF time was defined as time when medication has worn off and is no longer providing benefit with regard to mobility, slowness, and stiffness. ON time was defined as time when medication is providing benefit with regard to mobility, slowness, and stiffness In the Istradefylline studies, all OFF periods were included in the calculation of total OFF duration including OFF periods prior to taking the first morning dose of anti-PD medication.

The number of daily diaries used to calculate average daily OFF duration at baseline and follow up visits varied from 2 diaries in the North American studies and Study 6002-EU-007 to four diaries in the Japanese studies and 3 diaries for Study 6002-014. The degree to which a patients' clinical condition varies day to day, from relatively symptom free to more symptomatic with increased OFF time, was not discussed by the applicant. In the cited research paper from Hauser et al, further validating use of the home diary and its implementation in clinical trials, subjects completed daily diaries on 3 consecutive days. In this study the authors noted that 'reliability increased with increasing number of diary days but compliance diminished beyond 3 days'. (Hauser, Deckers, Lehert 'Parkinson's disease home diary: further validation and implications for clinical trials'. Mov Disord. 2004 Dec; 19(12):1409-13.)

In general, PD is commoner in males than in females and this was the case in all of the pivotal trials, except for those conducted in Japan where there was a higher proportion of females. Patients in the Japanese studies also had a lower BMI compared to patients in studies conducted outside of Japan. Background and concomitant levodopa dose in mg was also lower for Japanese patients compared to participants in the other six pivotal trials, however when adjusted for dose per kg this was less apparent.

The treatment duration in the pivotal studies was 12 weeks with the exception of Study 6002-EU-007 which had a 16-week treatment duration. There were three extension studies: one in North America which terminated early (Study 6002-US-007); a year-long extension of Study 6002-09 in Japan (Study 6002-010); and a similar length extension study of Study 6002-014 (Study 6002-018).

The primary endpoint for the pivotal studies was either change from baseline in duration of OFF time whilst awake, as recorded in the patient diary or change in the percentage of time spent in the OFF state whilst awake. In the case that percentage change from baseline was the primary endpoint, then change from baseline in the duration of time spent in the OFF state was a secondary endpoint and vice versa. In five of the 8 studies % change from baseline in OFF time was used and in the remaining three, change in total hours from baseline in OFF time was used. In the majority of studies change compared to baseline was measured at endpoint which was defined as Week 12 or the last visit by the patient.

A decrease in OFF time is not necessarily a therapeutic benefit as it can be associated with an increase in ON time with troublesome dyskinesia (not a good therapeutic outcome) and/or due to an increase in sleep time (neither a good nor bad therapeutic outcome). In order to conclude on treatment benefit, statistically significant changes in the OFF-time variable should be coupled with a statistically significant improvements in ON time without troublesome dyskinesia (i.e., ON without any dyskinesia + ON with non-troublesome dyskinesia). ON time without troublesome dyskinesia was the key secondary endpoint

with multiplicity adjustment, only in Study 6002-014; this endpoint was prospectively planned in the SAP of 6 trials (007, 013, 014, 018 and in the two Japanese trials 0608 and 009). Conversely, this endpoint was analysed *post hoc* in Studies 6002-US-005 and 6002-US-006, after unblinding of data.

The applicant conducted a pooled analysis of the five positive studies and of all eight pivotal studies. The primary endpoint in the pooled analysis was change from baseline in duration of OFF time. The fact that the studies lacked an identical common primary endpoint and did not adjust for multiplicity presents challenges to the interpretation of the results of the pooled analysis.

The most important secondary efficacy endpoint was change in baseline in time spent ON with various categories of dyskinesia, of which the most relevant is judged to be time ON without troublesome dyskinesia, which encompasses time ON without dyskinesia and time ON with non-troublesome dyskinesia. There were a number of other secondary endpoints including change from baseline in UPDRS Part II and Part III scores, CGI and other quality of life measures. All studies evaluated change in UPDRS II and III. Of note none of the studies made any adjustment for multiplicity of testing. This presents challenges in the interpretation of secondary endpoints and thus these can only be considered as descriptive and cannot provide substantial supportive evidence for the efficacy analysis.

Efficacy data and additional analyses

A number of Istradefylline posologies were evaluated in the pivotal studies from 10mg per day to 60mg day. The majority of studies investigated posologies of 20mg and/or 40mg per day.

The results from the eight pivotal studies are inconsistent. Efficacy was demonstrated in 4 out of 8 pivotal studies. There was demonstrated efficacy in 2 of the North American studies (6002-US-005 and 6002-US-013) and in both Japanese studies (6002-0608 and 6002-009). Study 6002-US-006, a North American study, very similar to Study 6002-US-005 except for istradefylline dose used, was a formally failed study. However, as argued by the applicant, it reached borderline significance when an ANCOVA analysis was used. Of most concern is that no evidence of efficacy was demonstrated in either study with European participants (Study 6002-EU-007 and Study 6002-014), nor in the North American study 6002-US-018.

The primary endpoint for the North American studies was decrease from baseline in percentage of awake time spent in the OFF state, whilst the primary endpoint for the Japanese studies was the decrease from baseline in the amount of awake time spent in the OFF state.

The two Phase 2b North American studies 6002-US-005 and 6002-US-006 were very similar in overall study design except that study 6002-US-005 explored a dose of 40mg istradefylline versus placebo and study US-006 explored two doses of istradefylline; 20mg and 60mg versus placebo.

In study 6002-US-005, the result of the primary endpoint analysis was statistically significant, with a reduction in the % of awake time spent in OFF state between baseline and endpoint -10.49% and -3.71% for istradefylline 40 mg/day and the placebo groups, respectively. The LSM difference from placebo in the change from baseline to endpoint in the percentage of awake time spent in the OFF state of -6.78% for the istradefylline 40 mg/day group was statistically significant (p=0.007, 95% CI -11.63,-1.92) by ANOVA. However it is noted the CIs were quite wide which adds some uncertainty.

In study 6002-US-006, the primary endpoint for % change from baseline to endpoint in time awake spent in OFF state was not statistically significant for istradefylline compared to placebo for the primary analysis method ANOVA pre-defined in the SAP for either the 20mg (-3.65%, p=0.088, 95% CI -7.83, 0.53) nor the 60mg (-3.77%, p=0.082, 95% CI -8.01, 0.47) istradefylline doses. The applicant repeated the analysis using ANCOVA which showed a marginally significant difference for the overall treatment effect compared to the placebo group (p-value = 0.049). In terms of effect size, with this additional

ANCOVA analysis, there was little difference noted between the 20mg and 60mg istradefylline doses raising further concern for the lack of support to recommend higher doses will provide additional clinical benefit. By the ANCOVA analysis, for the individual doses the LSM differences from placebo in the changes from baseline to endpoint in percentages of awake time per day spent in the OFF state were -4.35% and -4.49% for istradefylline 20 and 60 mg/day, respectively ([p=0.026, 95% CI -8.16, -0.54] and [p=0.024, 95% CI -8.35, -0.62]) respectively. Again the wide CIs add further clinical uncertainty. Treatment effect on ON time without troublesome dyskinesia was evaluated only in a *post hoc* analysis, and was not statistically significant either. In failing to meet the primary endpoint, by prespecified analysis method, Study US-006 is considered a formally failed study.

Study US-013 a Phase 3 study with similar inclusion criteria to the above two studies explored a single dose of 20mg istradefylline and results were statistically significant with the LSM difference from placebo in the changes from baseline to endpoint in the percentage of awake time per day spent in the OFF state was -4.57% (p=0.025, 95% CI -8.55, -0.59) for the istradefylline 20 mg/day group, by ANCOVA. However, this effect on the primary endpoint was not coupled with statistically significant differences in the main secondary endpoint, the percentage of awake time per day spent in an ON state without troublesome dyskinesia, casting doubts on the clinical relevance of the observed effect on the primary endpoint. As aforementioned, similar concerns are also raised regarding the wide with the potential effect of the 20mg dose leading to a reduction in OFF time from baseline compared to placebo, of as much as -8.55% but equally as little as -0.59%.

The fourth North American study US-018, a Phase 3 study, with similar design and patient population to the earlier studies, explored three istradefylline doses and also sought to perform a dose response analysis using doses 10mg, 20mg and 40mg istradefylline. This study failed to meet its primary endpoint with the LSM differences from placebo in the change from baseline to endpoint in the percentage of awake time per day spent in the OFF state were 1.79% (p=0.319, 95% Ci -1.73, 5.31), 1.50% (p=0.408, -2.05, 5.05), and -0.66% (p=0.714, -4.21, 2.88), for the istradefylline 10, 20, and 40 mg/day groups, respectively, by ANCOVA. In this study there was a numerical trend for an increase in time awake spent in OFF state for istradefylline 10mg and 20mg relative to placebo, which is of course considered clinically unfavourable. The applicant claims that these results were due to a larger than expected placebo effect seen in the placebo arm of Study 6002-US-018. However, whilst the CHMP can agree that the placebo response in US-018 was greater than what was observed in the other istradefylline studies, the placebo effect seen in Study 018 was still within the range found in the Stowe et al (2010) review in trials testing adjunct treatments to levodopa. Study 018 remains a negative study, showing no difference of istradefylline from PBO.

The two Japanese studies a phase 2b and phase 3 study were essentially duplicates of each other in terms of design, treatments (Istradefylline 20mg and 40mg) and population studied.

In Study 6002-0608 the LSM changes from Baseline at Endpoint in the total hours of awake time per day spent in the OFF state estimated based on a ANCOVA model were -0.66 hours (95% C.I -1.08 to -0.25 hours) for the placebo group, -1.31 hours (-1.73 to -0.89 hours) for the 20 mg/day KW-6002 group, and -1.58 hours (-1.99 to -1.17 hours) for the 40 mg/day KW-6002 group. The LSM difference from placebo was -0.65 hours (95% C.I -1.23 to -0.07 hours) for the 20 mg/day KW-6002 group and -0.92 hours (-1.49 to -0.35 hours) for the 40 mg/day KW-6002 group. This approximates to a difference from placebo of 39 minutes for Istradefylline 20mg and 55 minutes for Istradefylline 40mg.

In Study 6002-09 effect sizes were smaller in all treatment groups. The LSM change from baseline (95% CI) at the endpoint estimated based on the ANCOVA model (observed case analysis in the FAS) was −0.23 h (−0.62 to 0.16 h) for the placebo group, −0.99 h (−1.38 to −0.60 h) for the 20 mg/day of KW-6002 group, and −0.96 h (−1.35 to −0.58 h) for the 40 mg/day of KW-6002 group. The LSM difference from the placebo group (95% CI) at the final evaluation was −0.76 h (−1.30 to −0.22 h) for the 20

mg/day of KW-6002 group and −0.74 h (−1.27 to −0.20 h) for the 40 mg/day of KW-6002 group. This approximates to a difference from placebo of approximately 45 minutes for both Istradefylline treatment arms.

The effect size for Istradefylline 40mg compared to placebo in Study 6002-08 is broadly similar to that seen in the Opicapone registration studies. However, it should be borne in mind that both studies were conducted in Japan and had more female than male participant and mean BMI of participants was lower than one would expect in Europe.

In Study 6002-EU-007, Istradefylline 40mg failed to show any benefit in the only pivotal study conducted with an active comparator (entacapone 200mg daily). In fact, there was no difference between Istradefylline 40mg in percentage reduction of OFF from baseline at endpoint (-4.53% for placebo and -5.14% for Istradefylline 40mg) and placebo. The reduction from baseline for entacapone was slightly greater at -7.82%. The difference versus placebo was not statistically significant.

Neither was any efficacy demonstrated in the most recent Study 6002-014, conducted in maximally and optimally treated patients who had a history of levodopa induced dyskinesia, higher baseline doses of levodopa (mean dosage in excess of 800mg/day; minimum dose 400mg/day), and were on a range of adjunctive treatments). The LSM reduction in time in the OFF state from baseline to endpoint was -0.88 (95% CI -1.19, -1.58) hours for placebo, -1.2 (95% CI -1.52, -0.89) hours for Istradefylline 20mg and -1.15 (95% CI -1.46, -0.84) for Istradefylline 40mg. The difference in LSMs from placebo were -0.32 and -0.27 hours for Istradefylline 20mg and Istradefylline 40mg respectively, overall a reduction of 14 to 18 minutes per day.

According to the study protocols all patients should have had their levodopa treatment optimised prior to study enrolment. However, it is difficult to conclude that all patients in the study populations had received optimal treatment given that optimal treatment was not defined and that there was no lead in period in which treatment could have been assessed and optimised, meaning there may have been significant inter subject variability in baseline disease control prior to study entry. Whilst the Japanese studies conducted between 2007 and 2011 showed more robust efficacy results, again for reasons based on regional, gender and background treatment differences, whether these can demonstrate external validity for the currently proposed EU population is questioned. Finally, in the most recent study which, was conducted in a population with levodopa induced dyskinesia, of whom 39% were from the EU, neither Istradefylline 20mg nor Istradefylline 40mg showed any benefit over placebo, raising further questions about its efficacy in a maximally treated/more severe population.

Responses to D120 major objections

Four efficacy major objections were raised at D 120 that encompassed the following: (1) the applicant was asked to discuss the inconsistency of the results and justify how the evidence submitted (from the 8 trials) supports the claimed indication; (2) given the failure to demonstrate efficacy in studies with European populations the applicant was asked to provide a justification for the generalisability of studies with a positive result to the European population; (3) to justify the clinical relevance of the effect size and to perform responder analyses; and (4) to justify the proposed indication and the inclusion of patients with more severe PD in the indication.

Inconsistency in study results

The applicant considered that the inconsistencies seen in their study results are due to the inherent heterogeneity of PD in terms of disease progression and response, and the increased susceptibility of PD trial participants to demonstrate a placebo response. There is evidence that a placebo response may in some cases lead to increased production of endogenous dopamine from the striatum. However the applicant failed to provide any objective evidence to demonstrate that a heightened placebo effect was responsible for failure to show benefit in certain studies. In support of the claim regarding heterogeneity

the applicant submitted data from two systematic reviews by Stowe et al (2010) and Li (2017). However neither of these reviews demonstrated the same level of inconsistency of results as seen with the applicants pivotal trials.

Given the inconsistency of the individual study results the applicant undertook a pooled analysis of the 8 pivotal studies. This pooled analysis had not been predefined, but was performed as a rescue measure following the individual trials that were planned to stand on their own but showed only borderline or no effect. Initially a fixed effects model and later - given the heterogeneity of the studies considering their regional and wide temporal spread - a more appropriate random effects model was used. The estimated effect size with the random effects meta-analysis was modest and while the 95%-confidence interval excluded '0', In fact, and given the proximity of the CI-boundaries to 'no effect', lack of a treatment effect cannot be excluded. Besides this a larger sample size (as a consequence of meta-analytic pooling of different studies) allows smaller p-values in case of small effects, which would also not be convincing. It was considered that the standard pre-requesite, defined in EMA guidelines, of pooled 95% confidence interval well away from zero was not met. (CPMP/EWP/2330/99 -guideline about points to consider on applications with 1. Meta-analysis ; 2. One pivotal study).

Thus the issue regarding inconsistency of study results with 4 negative and 4 positive trials was not considered satisfactorily resolved and additional concerns regarding clinical relevance were further pursued in the D180LOI.

European population

To justify efficacy in the European population the applicant conducted a *post ho*c random effect meta-analysis of the two studies in which European populations were included (-007, -014) comparing outcomes between European (defined as patients from all European countries participating in the study except for Ukraine and Russia) and Rest of the World patient populations for differences in OFF time, ON time without troublesome dyskinesia and a responder analysis (for each of reduction in OFF time ≥ 1 hour, and for increase in ON time without troublesome dyskinesia ≥ 1 hour). Treatment effect sizes were greater in the European compared to the Rest of the World population. The effect size for reduction in OFF time in the European population with Istradefylline 20mg compared to placebo was -0.49 hours (95% CI: -1.2, 0.22) and for Istradefylline 40mg -0.39 hours (95% CI:-1.0, 0.22). This is similar to the effect seen in the Random effect meta-analysis for the 8 pivotal studies (-0.45 hours for Istradefylline 20mg and -0.46 for Istradefylline 40mg. However, the CIs are wide and include 0.

Regardless, the CHMP did not agree that exclusion of Ukraine from the EU population is necessarily justified. As such reviewing the original subgroup analysis by Region in the overall pool of 8 Phase 2b/3 studies showed that primary efficacy results in Europe and North America can be considered largely similar.

Conversely a larger effect was observed in Japan in comparison to Europe and North America. Several factors have been identified that may have contributed to the greater effect observed in the Japanese study; these factors relate to both difference in baseline characteristics of the enrolled population (M/F ratio, time in OFF state and modified H&Y stages at baseline), and to differences in study design (different number of valid diaries used for the endpoints definition at each assessment point) between Japanese trials and non-Japanese trials.

The issue of Studies 6002-EU-007 and 6002-014 being negative studies remains an unresolved issue and no demonstration of clinical benefit has been convincingly demonstrated in the EU population. A further MO in relation to efficacy in the European population was pursued was included in the D180LOI.

Clinical Relevance

The applicant has not satisfactorily addressed the issue of the clinical relevance of istradefylline as an additional therapy for patients with PD.

In order to conclude on treatment benefit, significant changes in the OFF-time variable have to be coupled with significant improvements in ON time without troublesome dyskinesia (i.e., ON without any dyskinesia + ON with non-troublesome dyskinesia). This was the rationale behind the request of a responder analysis combining in the responder definition at least 1 Hour Reduction in OFF Time plus 1 Hour Increase in ON Time without Troublesome Dyskinesia.

This responder analysis showed that in study 014 (conducted in subjects "optimally treated") the percentage of responders was higher in PBO (39%) that in the two istradefylline group (20 mg: 35%, 40 mg: 34%). This is a cause of concern as it suggests that in an optimally treated population there is no net benefit of adding istradefylline, but rather possibly a worse overall control of changes in OFF time coupled with good ON.

Furthermore this responders analysis shows a complete absence of effect in Study 018 and a very small difference from PBO in terms of percentage of responders (only 3% more responders with istradefylline than with PBO: 31% vs 28%) in Study EU-007.

Issue was not considered resolved and a further MO was included in the D180LOI.

Proposed Indication

The proposed indication adjunctive treatment to levodopa-based regimens in patients with PD experiencing "OFF" time in adults' was regarded as too broad as it could include patients experiencing other forms of OFF besides end of dose motor fluctuations. The applicant has proposed to limit the indication to those experiencing end of dose fluctuations in line with similar adjunctive PD products. The CHMP considers further revision to specify 'motor fluctuations' might also be more appropriate.

The applicant conducted another meta-analysis comparing a sub-group who would have met the key entry criteria for Study 6002-014 with those who would not have met the entry criteria with regard to change in OFF time from baseline and increase in ON time without troublesome dyskinesia. However patients from Study 6002-014 were not included in the analyses so even though the results appear favourable for use in a more severe population they are undermined by the exclusion of the study population from Study 6002-014 who failed to show any evidence of benefit from Istradefylline. Study 6002-014 was the trial performed upon FDA request to show that istradefylline treatment on top of optimized PD background therapy would result in added benefit for the patients. Although Study 6002-014 was the largest pivotal trial relative to size of each treatment group (nearly 200 patients per arm), this trial failed to show any evidence of efficacy for istradefylline.

It was not concluded that the efficacy in a maximally treated 'severe' PD population had been convincingly demonstrated, thus further MO was raised in relation to efficacy in the 'severe' population was included in the D180LOI.

Responses to Day 180 major objections

Following CHMP discussion, four major objections were raised at Day 180 relating to the clinical relevance of the effect size; efficacy in the "severe" population; efficacy in the European population and efficacy of the 40mg istradefylline dose compared to the 20mg dose.

Uncertainty of effect size and questionable clinical relevance

To demonstrate the clinical relevance of the effect size the applicant provided preliminary data from three patient surveys, two of which were commissioned by the applicant. The main study which was

quantitative in nature sought to investigate whether a decrease in OFF time of 20 or 30 minutes or an increase in ON time without troublesome dyskinesia would be regarded as beneficial by patients. The results of the study were supportive; however the patient population were not reflective of the patient populations in the pivotal studies, with 60% of survey participants spending 1 hour or less in the OFF state daily compared to a range of a mean of 5.36 hours/day to 6.7 hours/day for patients in the pivotal studies. Therefore the survey results cannot necessarily be extrapolated to the populations in the pivotal studies.

Further analyses provided by the applicant for 'response', defined as a reduction in OFF time ≥1 hour from baseline and/or an increase in ON time without troublesome dyskinesia of ≥1 hour from baseline, or reduction in UPDRS III score ≥30% from baseline, evaluating the number of responses recorded across the study from Week 2 to 12 did not provide any additional evidence to support the clinical relevance of the effect sizes seen in the pooled pivotal studies nor did it diminish the uncertainty with regard to the true pooled effect sizes for reduction in OFF time from baseline compared to placebo or increase in ON time without troublesome dyskinesia from baseline compared to placebo. Issue was not considered resolved

Efficacy in the severe population

The applicant has repeated the sub-group analysis comparing all of those who would have met the 3 key inclusion criteria for Study 6002-014 with the study population of Study 6002-014 included, with those who would not have met the criteria. Reduction in OFF time (Istradefylline 20mg: -0.35 hours v -0.41 hours; Istradefylline 40mg: -0.32 hours v -0.45 hours) and increase in ON time without troublesome dyskinesia (Istradefylline 20mg: 0.32 hours v 0.43 hours; Istradefylline 40mg 0.25 hours v 0.33 hours) were both slightly lower for subjects who met the 3 key criteria (i.e those who would have met the criteria for inclusion in Study 014) in both Istradefylline treatment arms compared to those who did not.

A further sub-group analysis using surrogates for severity derived from the literature and expert opinion was also performed showed that in general overall change from baseline in OFF time, ON time without troublesome dyskinesia and UPDRSIII score did not appear worse in more "severe" patients. In the subgroups according to total hours OFF time, a greater istradefylline effect (both for 20 mg/ day and for 40 mg/day) was observed in subjects with the higher number total hours per day in the OFF state (3rd tertile, corresponding to ≥7.1 hours OFF time). The total daily number of hours spent in the OFF state appears to be a treatment effect modifier and is the only factor shown to be so at both dose levels. This was confirmed by the multivariate regression analyses performed by the applicant, which showed only gender, age at onset, and baseline OFF time to be independent modifiers of the OFF time treatment effect in istradefylline studies at both dose levels.

The applicant also performed an additional post hoc 'Number of Response Timepoints' analysis. Again, this analysis whilst appearing to support a trend for benefit for istradefylline in OFF time, ON time and UPDRSIII score, remains exploratory and does not overcome the formally failed primary endpoint. The applicant argues that these patterns in the data may occur as a result of heterogeneity in PD and exemplify the limitations of assessing benefit based on outcome at a single timepoint. However these results bring into question the possible absence of persistence of the effect, also considering that according to the "Guideline on clinical investigation of medicinal products in the treatment of Parkinson's disease" study duration should be at least 3 months; thus earlier timepoints may be considered less relevant. As for any post hoc analysis- a number of choices are taken that may affect final results. As commented in the previous round of the procedure, these post hoc analyses cannot compensate for the failure of the pre-planned Study-6002-014.

Overall it appears that study participants who could be described as maximally treated had slightly poorer outcomes than other study participants. Bearing in mind the limitations of post hoc analysis there is some evidence that patients who could be defined as having more severe PD have similar outcomes than

the non-severe population. In addition the total daily OFF time appears to be a treatment effect modifier at both dose levels. Issue was not considered resolved.

Efficacy in the European population

The applicant has re-analysed Pool E1 data by region with Ukraine included as part of Europe. Numerically, the effect observed on OFF time in the European subgroup was similar to the effect observed in the overall population (but lower than that seen in the Japanese population); given that the confidence interval of effect in the European subgroup included the absence of effect and given that the two studies that enrolled the European population (Study EU-007 and Study 014) were both negative studies according to pre-specified primary analyses, it may be argued that the statistically significant results in the overall population combining the results of all 8 Phase 2b/Phase 3 studies through meta-analysis is likely a consequence of sample size increase, favouring statistical significance of small effects.

It is unfortunate that the two studies conducted in European populations both did not meet their primary endpoint. These were both international studies and potentially the results may have been negatively impacted by the 'rest of world' population whom appeared to have worse outcomes compared to the overall population.

Therefore, it remains inconclusive whether the effect in European patients is markedly different to the overall effect of istradefylline across the development programme. The inconsistency in the results of the 8 pivotal trials conducted in different populations leads to considerable uncertainty in the true effect of istradefylline.

Efficacy of the 40mg dose

The applicant proposes in the SmPC that a starting dose of 20mg should be used which can be increased after 4 weeks to 40mg if there is clinical need. However there is no evidence of greater efficacy of the 40mg dose compared to the 20mg dose.

The applicant performed a further post hoc meta-analysis of 4 studies in which both the 20mg and 40mg Istradefylline doses were evaluated versus placebo. For the primary and key secondary endpoints, there was no difference in LSM change from baseline at 12 weeks for OFF Time, nor ON time without troublesome dyskinesia between the 20mg and 40mg Istradefylline doses. Istradefylline 40mg showed a reduction in UPDRS III score of 0.8 compared to Istradefylline 20mg. The clinical relevance of a decrease of this size in the UPDR III score is questioned. In addition it was not possible to identify any subgroups who might be more likely to benefit from the 40mg dose than the 20mg dose. Overall Istradefylline displays a lack of dose dependency between the 20mg and 40mg doses and from an efficacy point of view the wording in section 4.2 of the SmPC regarding the 40mg dose cannot be supported. In addition, the concern remains that there is overall inconsistency in the istradefylline clinical development programme, making it difficult to conclude on the benefit of either the 20mg or 40mg doses. Issue was not considered resolved.

Oral explanation at CHMP

The applicant was invited to attend an oral explanation at the meeting of the CHMP on June 23rd, 2021 to address the remaining major objections, in particular as to whether efficacy had been demonstrated.

The applicant further defend the pooling strategy as an appropriate way to evaluate data in the istradefylline programme. However, the CHMP is of the opinion that the inconsistency of study results, including the different responses observed in different populations and the difference in response over time, are not resolved by *post hoc* pooling the individual trials intended to rescue their unconvincing results. In addition, the estimated modest effect size, which is accompanied by an unadjusted nominal confidence interval in close proximity to no effect, and the observation of no clear pattern of a dose

response with increasing doses of istradefylline, leads to considerable uncertainty on the effect of the treatment and its clinical relevance.

As per results of studies including EU populations, the applicant claimed a lack of assay sensitivity for study 6002-EU-007. In that study, the only one conducted with an active comparator (entacapone 200mg daily), there was no difference between Istradefylline 40mg in percentage reduction of OFF from baseline at endpoint (-4.53% for placebo and -5.14% for Istradefylline 40mg) and placebo. While the reduction from baseline for entacapone was slightly greater at -7.82%, it can be agreed that the difference versus placebo was not statistically significant. They presented the argument that trials in PD present intrinsic difficulties and heterogeneous outcomes are expected. This argument is not accepted, as whilst it is agreed that studies conducted in Parkinsons disease have demonstrated some heterogeneity, the heterogeneity and inconsistency observed in the istradefylline development programme exceeds that of the meta-analysis presented by the applicant from Stowe et al. It should also be noted that this MAA assess the B/R for Istradefylline specifically and the applicant is expected to plan studies adequately taking into account any potential heterogeneity. Additionally, the applicant presented a random effect meta-analisis for change from baseline in total hours per day in OFF State at 12 Weeks (Europe vs Non-Europe, Studies 6002-EU-007 and -6002-014) and concluded that European patients did not contribute to lack of assay sensitivity. Differences between istradefylline 20mg / 40mg versus placebo included the null value. The applicant did not discuss the reasons why Study 6002-014 conducted in 'maximally and optimally treated' patients did not show positive results.

In order to further defend the clinical relevance of istradefylline treatment effect, the applicant further discussed the results from three patient surveys, two of which were commissioned by the applicant. The main result was that nearly 80% of patients would find beneficial getting " 20 minutes less OFF time with very few additional side effects". Further, the applicant claimed that "*neurology experts practising in Europe have also conveyed the results are representative of the sentiments expressed by patients in the clinic*". The results could be considered as supportive but CHMP highlighted that the study population included in the surveys was clearly different in terms of average daily OFF state from the one included in the istradefylline trials (see above discussion). This aspect was acknowledged by the applicant who noted that it was intended to recruit a broader PD population in the surveys. Therefore, the survey results cannot necessarily be extrapolated to the populations in the pivotal studies. It should be noted that the question posed to the participant made reference to "*few additional side effects*". As such, the CHMP highlighted that even if it can be agreed that the safety profile has been well-characterized, several ADRs were identified and that there are concerns that the higher proposed dose of istradefylline (40 mg/day) could be slightly less safe as compared to the lower dose especially in relation to the development of serious and severe TEAEs after a long term treatment with istradefylline. Finally, CHMP noted that 20-30 minutes of reduction of OFF time is a result derived from an analysis that was not considered adequate (see above).

Finally, the applicant presented a meta-analysis of 4 pivotal studies included a direct comparison of both 20 mg/day and 40 mg/day doses to further defend the proposed posology. Additionally, the applicant emphasised the need for an individualised PD therapeutic regiment. The CHMP acknoledged that PD therapeutic management may benefit from an individualised approach but highlighted the lack of evidence for the added benefit of Istradefylline 40mg over that of Istradefylline 20mg across 8 pivotal studies and the potential for increased serious and severe TEAES with the 40mg dose during long-term treatment.

The CHMP concluded following the oral explanation and discussion that efficacy had not been demonstrated and that the benefit risk balance for Istradefylline was negative.

2.5.3. Conclusions on the clinical efficacy

Overall the studies have produced inconsistent results with efficacy demonstrated in 2 of the older North American studies and in two Japanese studies. The effect sizes are variable, and clinical relevance is questioned for any effects that have been observed.

It may be argued that the statistically significant results in the overall population combining the results of all 8 Phase 2b/Phase 3 studies through a *post hoc* meta-analysis is likely a consequence of sample size increase and possibly driven by greater efficacy in the Japanese populations.

Even if the justification for inconsistency in study results regarding heterogeneity in PD and potential for an enhanced placebo effect were accepted, the random effects meta-analysis of the eight pivotal studies for change from baseline in OFF time and change in ON time without troublesome dyskinesia are extremely modest with large CIs with the lower confidence interval close to zero, which suggests that there is considerable uncertainty around the true size of the effects. Given the proximity of the CI-boundaries to 'no effect' a lack of a treatment effect cannot be excluded. In addition there appears to be no difference in efficacy between the istradefylline 20mg and 40mg/ day doses. The clinical relevance of the effect sizes seen for both doses of Istradefylline is questioned. The responder analyses have not provided any convincing evidence to support a clinical benefit of istradefylline that is considerably better than placebo.

Istradefylline displays a lack of dose dependency between the 20mg and 40mg doses. In addition, the concern remains that there is overall inconsistency in the istradefylline clinical development programme, making it difficult to conclude on the benefit of either the 20mg or 40mg doses.

Overall, the applicant has not satisfactorily addressed all the issues raised as Major Objections and the available data remain inconsistent and unconvincing overall clinical efficacy has not been demonstrated.

2.6. Clinical safety

Patient exposure

Safety data pooling strategy:

Istradefylline safety has been evaluated in 52 clinical trials in which 4323 subjects were exposed to istradefylline. In addition to clinical trial experience, as of 31 May 2019 approximately 63,500 patients have been exposed to istradefylline in the post-marketing setting in Japan.

Patients were exposed to istradefylline in the following trials:

- 822 subjects were exposed to istradefylline in twenty-eight Phase 1 studies
- 3501 subjects were exposed to istradefylline in Twenty-four Phase 2/3 studies and this safety data were organized into 5 pools:

Pools 1 and 2, which include double-blind placebo-controlled studies and open label long-term treatment experience for the indication sought, respectively, are the most relevant for evaluating safety associated with the proposed indication.

Pools 3, 4, and 4a provide supportive information on istradefylline safety. Pool 3 includes Phase 2 pilot studies in PD. Pool 4 includes data from studies of other indications and Pool 4a includes data from 2 clinical studies of istradefylline as monotherapy in PD.

Table 98: Organization of Pooled Data for the 24 Phase 2 and 3 Studies Included in the Summary of Clinical Safety

Pool 1	Pool 2	Pool 3	Pool 4	Pool 4a
Double-blind, randomized, placebo-controlled, fixed-dose studies Subjects with PD, treated with levodopa/carbidopa or levodopa/benserazide Doses: 10, 20, 40, 60 mg/day Treatment duration: 12 or 16 weeks Placebo = 1010 Istradefylline = 2073	Open-label, long-term studies[a] Subjects with PD, treated with levodopa/carbidopa or levodopa/benserazide Doses: 20, 40, 60 mg/day Treatment duration: 52 or 104 weeks[b] Istradefylline = 2132[c] Prior istradefylline = 1343 No prior istradefylline = 789	Phase 2 pilot studies Subjects with PD Doses: 5, 10, 20, 40, 80 mg/day and single doses of 50, 100, 200, and 300 mg Treatment duration: single dose (up to 4, each) Placebo = 67 Istradefylline = 149	Other Indications Doses: 40, 80, 120 mg/day Treatment duration: to 12 weeks[d] Placebo = 329 Istradefylline = 490	Studies as monotherapy in PD Dose: 40 mg/day Treatment duration: to 12 weeks[d] Placebo = 82 Istradefylline = 164
Studies (8): 6002-US-005 6002-US-006 6002-US-013 6002-US-018 6002-EU-007 6002-0608 6002-009 6002-014	Studies (5): 6002-US-007 6002-INT-001 6002-US-025 6002-010 6002-018	Studies (5): 6002-US-001 6002-US-004 6002-EU04 6002-EU05 6002-0406	Studies (6): Major depressive disorder (N=361) 6002-US-101 6002-US-104 Restless legs syndrome (N=212) 6002-US-012 6002-US-201 Monotherapy in PD (N=218) 6002-0407[d] 6002-US-051	Studies (2) 6002-0407[d] 6002-US-051

- *Pool 1 (DB, Placebo-controlled, Fixed-dose Studies)*

Pool 1 includes 8 DB, randomized, placebo-controlled fixed-dose studies of 12- or 16-weeks duration. In Pool 1-studies, subjects had idiopathic PD and motor response complications with levodopa (i.e., the patient population targeted in the proposed indication).

All subjects were receiving levodopa, in combination with a peripheral dopa-decarboxylase inhibitor (carbidopa or benserazide), and most were also receiving other additional standard anti-parkinson medication(s). In Pool 1 2073 patients were exposed to istradefylline

Most of the subjects (1765 of 2073 subjects) in Pool 1 who received istradefylline were treated with the proposed dosing regimen of 20 or 40 mg/day.

- *Pool 2 (OL, Long-term Studies)*

Pool 2 includes 5 OL, long-term extension studies of istradefylline in subjects with idiopathic PD and motor response complications while taking levodopa therapy. Eligible subjects who completed treatment with the investigational product (istradefylline, entacapone, or placebo) in short-term (12- or 16-week) DB, placebo-controlled studies were allowed to enter the long-term studies. The 5 long-term studies

comprising Pool 2 provide safety data from a large number of istradefylline-treated subjects (i.e., 2132 subjects with PD, including 1345 subjects treated with istradefylline for at least 1 year.

- *Pool 3 (Phase 2 Pilot Studies)*

Pool 3 combines the safety data of istradefylline in subjects with idiopathic PD enrolled in the smaller Phase 2 studies conducted earlier in the development program to test proof of concept and identify a dose. In this pool only 149 patients were exposed istradefylline.

- *Pool 4 (Other Indications)*

Pool 4 combines the safety data from 6 clinical studies of istradefylline as monotherapy in PD (2 studies), major depressive disorder (2 studies), and RLS (2 studies). In this pool 490 patients were treated with istradefylline.

- *Pool 4a (Monotherapy in PD, subset of pool 4)*

Pool 4a combines the safety data from 2 clinical studies of istradefylline as monotherapy in PD. In these two studies 164 patients received the istradefylline treatment.

Overall Extent of Exposure

A total of 822 subjects were exposed to at least 1 dose of istradefylline in a Phase 1 study; most of the subjects who participated in a Phase 1 study were exposed to istradefylline for 1 to 3 days (52%; 430/822 subjects).

Exposure in Phase 2 and 3 Studies

Overall, 3501 subjects took at least 1 dose of istradefylline in a Phase 2 or 3 study.

The total exposure to istradefylline for subjects in a Phase 2 or 3 study was 3079 PYs; 86% of these subjects (2997/3501 subjects; 3021 PYs) received istradefylline as adjunctive therapy for PD. A total of 1215 subjects were exposed to istradefylline for longer than 12 months and 413 subjects were exposed to istradefylline for longer than 24 months as adjunctive therapy for PD. In the Phase 2 and 3 studies of istradefylline administered as adjunctive therapy in PD 52% were between 18 and 64 years of age (1556/2997 subjects), 37% were between 65 and 74 years of age (1098/2997), 11% were between 75 and 84 years of age (332/2997), and only 0.4% (11) were ≥85 years old. This last age category was under-represented.

Table 99: Exposure to Istradefylline in Phase 2 and 3 Studies

Duration of Therapy[a]	Number of Subjects	Exposure (patient-years)
Cumulative for All Indications		
≤ 3 months (13 weeks)	1447	230.0
> 3 months to ≤ 6 months (26 weeks)	292	105.2
> 6 months to ≤ 12 months (52 weeks)	541	454.4
> 12 months to ≤ 18 months (78 weeks)	732	888.5
> 18 months to ≤ 24 months (104 weeks)	70	122.8
> 24 months	413	1278.7
Missing	6	-
Total	3501	3079.4
Cumulative for Adjunctive Therapy in PD		
≤ 3 months (13 weeks)	951	171.6
> 3 months to ≤ 6 months (26 weeks)	290	104.6
> 6 months to ≤ 12 months (52 weeks)	541	454.4
> 12 months to ≤ 18 months (78 weeks)	732	888.5
> 18 months to ≤ 24 months (104 weeks)	70	122.8
> 24 months	413	1278.7
Total for Adjunctive Therapy in PD	2997	3020.5
Cumulative for Monotherapy in PD		
≤ 3 months (13 weeks)	176	26.5
> 3 months to ≤ 6 months (26 weeks)	2	0.5
Total for Monotherapy in PD	178	27.0
Cumulative for RLS		
≤ 3 months (13 weeks)	104	10.0
Total for RLS	104	10.0
Cumulative for MDD		
≤ 3 months (13 weeks)	216	21.9
Missing	6	-
Total for MDD	222	21.9

Exposure to Istradefylline by Dose in Phase 2 and 3 Studies

The majority of subjects in the Phase 2 and 3 studies received 40 mg/day of istradefylline (63%; 2206/3501 subjects)

Table 100: Exposure to Istradefylline by Dose in Phase 2 and 3 Studies

Istradefylline dose	Number of Subjects	Exposure (patient-years)
Cumulative for All Indications		
10 mg/day	42	10.1
20 mg/day	894	605.1
40 mg/day	2206	2333.6
50 mg/day	3	0.0
60 mg/day	192	115.3
≥ 80 mg/day	164	15.3
Total	3501	3079.4
Cumulative for Adjunctive Therapy in PD		
10 mg/day	42	10.1
20 mg/day	880	603.5
40 mg/day	1862	2290.0
50 mg/day	3	0.0
60 mg/day	192	115.3
≥ 80 mg/day	18	1.6
Total for Adjunctive Therapy in PD	2997	3020.5
Cumulative for Monotherapy in PD		
20 mg/day	14	1.5
40 mg/day	164	25.4
Total for Monotherapy in PD	178	27.0
Cumulative for RLS		
40 mg/day	99	9.5
≥ 80 mg/day	5	0.5
Total for RLS	104	10.0
Cumulative for MDD		
40 mg/day	81	8.6
≥ 80 mg/day	141	13.3
Total for MDD	222	21.9

Adverse events

Treatment-emergent adverse events (TEAEs)

General information

Pool 1 (DB, Placebo-controlled, Fixed-dose Studies)

Seven of the studies included in Pool 1 were 12 weeks in duration and 1 study was 16 weeks in duration. Overall, 72.4% of subjects in the total istradefylline group experienced at least 1 TEAE compared to 65.4% of subjects in the placebo group. The incidences of subjects with any TEAE were similar in the istradefylline 20 mg/day and 40 mg/day groups (70.7% and 70.1% respectively), and higher in the istradefylline 10 mg/day and 60 mg/day group (82.4% and 85.8% respectively). Of note, the number of subjects in the 10 mg/day and 60 mg/day groups were much smaller than the 20 mg/day and 40 mg/day.

The 10 mg/day and 60 mg/day doses were only used in 1 study each. The incidence of subjects with serious TEAEs considered related to study drug (i.e., definitely, probably, or possibly related to study drug) was 1.4% in the total istradefylline group and 0.9% in the placebo group with similar incidences in the istradefylline 20 mg/day (1.5%) and 40 mg/day (1.6%) groups.

Nine subjects (4 in the total istradefylline group [0.2%] and 5 in the placebo group [0.5%]) in Pool 1 had a TEAE with an outcome of death. No death was assessed as related to istradefylline treatment; for 3 placebo-treated subjects, the death was assessed as possibly or probably related to treatment.

TEAEs leading to discontinuation were reported in 6.5% of subjects in the total istradefylline group and 5.2% of subjects in the placebo group, with the greatest proportion reported in the istradefylline 60 mg/day group (10.3%).

Table 101: Overview of TEAEs: Pool 1

		Istradefylline				
Subjects with:	Placebo N=1010 n (%)	10 mg/day N=153 n (%)	20 mg/day N=869 n (%)	40 mg/day N=896 n (%)	60 mg/day N=155 n (%)	Total N=2073 n (%)
Any TEAE	661 (65.4)	126 (82.4)	614 (70.7)	628 (70.1)	133 (85.8)	1501 (72.4)
Any Serious TEAE	31 (3.1)	6 (3.9)	34 (3.9)	43 (4.8)	4 (2.6)	87 (4.2)
Any TEAE leading to discontinuation	53 (5.2)	5 (3.3)	49 (5.6)	65 (7.3)	16 (10.3)	135 (6.5)
Any related[a] TEAE	417 (41.3)	93 (60.8)	393 (45.2)	423 (47.2)	98 (63.2)	1007 (48.6)
Any related[a] serious TEAE	9 (0.9)	3 (2.0)	13 (1.5)	14 (1.6)	0	30 (1.4)
Any severe TEAE	67 (6.6)	18 (11.8)	66 (7.6)	71 (7.9)	19 (12.3)	174 (8.4)
Deaths	5 (0.5)	1 (0.7)	0	3 (0.3)	0	4 (0.2)

Pool 2 (OL, Long-term Studies)

In Pool 2 where the mean treatment duration was 69.61 weeks (median, 54.07 weeks, Section 2.7.4.1.2.4), 63.1% of subjects received istradefylline for 12 months or longer. Overall, 89.7% of subjects reported a TEAE (Table 2.7.4-13). Serious TEAEs were reported in 22.6% of subjects, and 6.0% of subjects reported a serious TEAE that was considered to be related to istradefylline treatment.

There were 32 deaths reported in Pool 2 (1.5%). Six deaths were assessed as related to study drug (istradefylline) by the investigators. However, all 6 subjects had significant confounding medical history and/or concurrent medical conditions.

TEAEs leading to treatment discontinuation were reported by 14.3% of subjects in Pool 2.

Table 102: Overview of TEAEs: Pool 2

Subjects With:	Total Istradefylline (20-60 mg/day) N=2132 n (%)
Any TEAE	1913 (89.7)
Any serious TEAE	482 (22.6)
Any TEAE leading to discontinuation	304 (14.3)
Any related[a] TEAE	1502 (70.5)
Any related[a] serious TEAE	127 (6.0)
Any severe TEAE	558 (26.2)
Deaths	32 (1.5)

Treatment-emergent Adverse Events by System Organ Class

Pool 1 (DB, Placebo-controlled, Fixed-dose Studies)

The System Organ Class (SOC) with the most frequently reported TEAEs was nervous system disorders, for which events were reported for 34.1% of subjects in the total istradefylline group and 26.8% of subjects in the placebo group with similar incidences reported for the 20 mg/day (32.0%) and 40 mg/day (31.7%) istradefylline groups. Other SOCs with TEAEs reported with at least a 10% incidence in the total istradefylline group were gastrointestinal disorders, psychiatric disorders, investigations, musculoskeletal and connective tissue disorders, and infections and infestations; all of which were reported with < 5% difference in the incidence between the total istradefylline and placebo groups.

Table 103: TEAS by SOC: Pool 1

		Istradefylline				
	Placebo N=1010 n (%)	10 mg/day N=153 n (%)	20 mg/day N=869 n (%)	40 mg/day N=896 n (%)	60 mg/day N=155 n (%)	Total N=2073 n (%)
Subjects with any TEAE	661 (65.4)	126 (82.4)	614 (70.7)	628 (70.1)	133 (85.8)	1501 (72.4)
System Organ Class [a]						
Nervous system disorders	271 (26.8)	73 (47.7)	278 (32.0)	284 (31.7)	72 (46.5)	707 (34.1)
Gastrointestinal disorders	151 (15.0)	36 (23.5)	145 (16.7)	179 (20.0)	48 (31.0)	408 (19.7)
Psychiatric disorders	115 (11.4)	25 (16.3)	127 (14.6)	146 (16.3)	38 (24.5)	336 (16.2)
Investigations	151 (15.0)	12 (7.8)	125 (14.4)	137 (15.3)	20 (12.9)	294 (14.2)
Musculoskeletal and connective tissue disorders	146 (14.5)	37 (24.2)	100 (11.5)	121 (13.5)	31 (20.0)	289 (13.9)
Infections and infestations	147 (14.6)	23 (15.0)	115 (13.2)	128 (14.3)	20 (12.9)	286 (13.8)
Injury, poisoning and procedural complications	91 (9.0)	18 (11.9)	84 (9.7)	82 (9.2)	15 (9.7)	199 (9.6)
General disorders and administration site conditions	91 (9.0)	16 (10.5)	69 (7.9)	76 (8.5)	17 (11.0)	178 (8.6)
Skin and subcutaneous tissue disorders	50 (5.0)	7 (4.6)	49 (5.6)	49 (5.5)	11 (7.1)	116 (5.6)
Respiratory, thoracic and mediastinal disorders	40 (4.0)	6 (3.9)	50 (5.8)	43 (4.8)	8 (5.2)	107 (5.2)
Vascular disorders	36 (3.6)	7 (4.6)	40 (4.6)	27 (3.0)	3 (1.9)	77 (3.7)
Metabolism and nutrition disorders	23 (2.3)	7 (4.6)	32 (3.7)	25 (2.8)	6 (3.9)	70 (3.4)
Cardiac disorders	35 (3.5)	3 (2.0)	26 (3.0)	25 (2.8)	8 (5.2)	62 (3.0)
Renal and urinary disorders	24 (2.4)	6 (3.9)	15 (1.7)	26 (2.9)	6 (3.9)	53 (2.6)
Ear and labyrinth disorders	12 (1.2)	3 (2.0)	15 (1.7)	20 (2.2)	1 (0.6)	39 (1.9)
Eye disorders	17 (1.7)	4 (2.6)	15 (1.7)	13 (1.5)	6 (3.9)	38 (1.8)
Blood and lymphatic system disorders	12 (1.2)	0	9 (1.0)	11 (1.2)	0	20 (1.0)
Reproductive system and breast disorders	8 (0.8)	3 (2.0)	10 (1.2)	4 (0.4)	0	17 (0.8)
Neoplasms benign, malignant and unspecified (incl. cysts and polyps)	7 (0.7)	0	5 (0.6)	8 (0.9)	3 (1.9)	16 (0.8)

		Istradefylline				
	Placebo N=1010 n (%)	10 mg/day N=153 n (%)	20 mg/day N=869 n (%)	40 mg/day N=896 n (%)	60 mg/day N=155 n (%)	Total N=2073 n (%)
System Organ Class [a]						
Surgical and medical procedures	2 (0.2)	0	5 (0.6)	1 (0.1)	2 (1.3)	8 (0.4)
Hepatobiliary disorders	3 (0.3)	0	1 (0.1)	4 (0.4)	1 (0.6)	6 (0.3)
Immune system disorders	1 (0.1)	0	1 (0.1)	2 (0.2)	2 (1.3)	5 (0.2)
Endocrine disorders	2 (0.2)	1 (0.7)	2 (0.2)	2 (0.2)	0	5 (0.2)
Social circumstances	0	0	0	0	1 (0.6)	1 (<0.1)

Pool 2 (OL, Long-term Studies)

The 10 most frequently reported SOCs with TEAEs in Pool 2 were the same as those reported for Pool 1 and Pool 2 (see the table below). This is not unexpected as 63.0% (1343/2132) of subjects in Pool 2 received istradefylline in Pool 1 and Pool 2 TEAEs reflect subjects' total istradefylline exposure, including events reported during short-term exposure to istradefylline in Pool 1 studies.

In Pool 2, the SOCs with TEAEs reported for ≥ 40% of subjects were nervous system disorders, psychiatric disorders, and gastrointestinal disorders.

Table 104: TEAS by SOC: Pool 2

	Total Istradefylline (20-60 mg/day) N=2132 n (%)
Subjects with any TEAE	1913 (89.7)
System Organ Class[a]	
Nervous system disorders	1374 (64.4)
Psychiatric disorders	924 (43.3)
Gastrointestinal disorders	886 (40.6)
Musculoskeletal and connective tissue disorders	835 (39.2)
Infections and infestations	793 (37.2)
Injury, poisoning and procedural complications	622 (29.2)
General disorders and administration site conditions	550 (25.8)
Investigations	544 (25.5)
Skin and subcutaneous tissue disorders	333 (15.6)
Respiratory, thoracic and mediastinal disorders	330 (15.5)
Renal and urinary disorders	241 (11.3)
Vascular disorders	222 (10.4)
Eye disorders	196 (9.2)
Metabolism and nutrition disorders	202 (9.5)
Cardiac disorders	172 (8.1)
Neoplasms benign, malignant and unspecified (incl. cysts and polyps)	113 (5.3)
Ear and labyrinth disorders	110 (5.2)
Reproductive system and breast disorders	108 (5.1)
Blood and lymphatic system disorders	79 (3.7)
Surgical and medical procedures	25 (1.2)
Hepatobiliary disorders	25 (1.2)
Endocrine disorders	23 (1.1)
Immune system disorders	30 (1.4)
Congenital, familial and genetic disorders	8 (0.4)
Social circumstances	3 (0.1)
Product issues	3 (0.1)

Treatment-emergent AEs by PT

Pool 1 (DB, Placebo-controlled, Fixed-dose Studies)

Treatment-emergent AEs by PT that were reported for ≥ 1.0% of subjects in the total istradefylline group are summarized in descending frequency for the total istradefylline group are presented in the table below.

The most frequently reported TEAEs for subjects in the istradefylline groups were dyskinesia, nausea, dizziness, constipation, fall, and insomnia. The incidences of dyskinesia, nausea, and dizziness in the istradefylline 60 mg/day group were greater than in the 20 and 40 mg/day groups.

Table 105: TEAE Reported for ≥ 1.0% of Subjects in the Total Istradefylline Group by Preferred Term: Pool 1

Preferred Term[a]	Placebo N=1010 n (%)	Istradefylline 10 mg/day N=153 n (%)	Istradefylline 20 mg/day N=869 n (%)	Istradefylline 40 mg/day N=896 n (%)	Istradefylline 60 mg/day N=155 n (%)	Istradefylline Total N=2073 n (%)
Subjects with any TEAE	661 (65.4)	126 (82.4)	614 (70.7)	628 (70.1)	133 (85.8)	1501 (72.4)
Dyskinesia	97 (9.6)	33 (21.6)	140 (16.1)	159 (17.7)	37 (23.9)	369 (17.8)
Nausea	46 (4.6)	11 (7.2)	52 (6.0)	54 (6.0)	35 (22.6)	152 (7.3)
Dizziness	42 (4.2)	8 (5.2)	44 (5.1)	44 (4.9)	22 (14.2)	118 (5.7)
Constipation	33 (3.3)	10 (6.5)	50 (5.8)	48 (5.4)	3 (1.9)	111 (5.4)
Fall	50 (5.0)	9 (5.9)	35 (4.0)	45 (5.0)	7 (4.5)	96 (4.6)
Insomnia	42 (4.2)	10 (6.5)	31 (3.6)	48 (5.4)	5 (3.2)	94 (4.5)
Parkinson's disease[b]	37 (3.7)	17 (11.1)	31 (3.6)	25 (2.8)	5 (3.2)	78 (3.8)
Viral upper respiratory tract infection	34 (3.4)	5 (3.3)	35 (4.0)	31 (3.5)	3 (1.9)	74 (3.6)
Headache	30 (3.0)	7 (4.6)	24 (2.8)	31 (3.5)	8 (5.2)	70 (3.4)
Back pain	29 (2.9)	6 (3.9)	30 (3.5)	24 (2.7)	10 (6.5)	70 (3.4)
Arthralgia	29 (2.9)	9 (5.9)	19 (2.2)	21 (2.3)	12 (7.7)	61 (2.9)
Hallucination	18 (1.8)	2 (1.3)	18 (2.1)	26 (2.9)	9 (5.8)	55 (2.7)
Somnolence	32 (3.2)	5 (3.3)	28 (3.2)	17 (1.9)	4 (2.6)	54 (2.6)
Anxiety	20 (2.0)	5 (3.3)	16 (1.8)	22 (2.5)	6 (3.9)	49 (2.4)
Blood creatine phosphokinase increased	27 (2.7)	1 (0.7)	21 (2.4)	21 (2.3)	6 (3.9)	49 (2.4)
Fatigue	19 (1.9)	6 (3.9)	18 (2.1)	20 (2.2)	4 (2.6)	48 (2.3)
Tremor	26 (2.6)	6 (3.9)	16 (1.8)	21 (2.3)	2 (1.3)	45 (2.2)
Diarrhoea	30 (3.0)	3 (2.0)	10 (1.2)	23 (2.6)	6 (3.9)	42 (2.0)
Urinary tract infection	20 (2.0)	5 (3.3)	16 (1.8)	19 (2.1)	2 (1.3)	42 (2.0)
Weight decreased	17 (1.7)	4 (2.6)	16 (1.8)	20 (2.2)	1 (0.6)	41 (2.0)
Upper respiratory tract infection	21 (2.1)	4 (2.6)	12 (1.4)	18 (2.0)	6 (3.9)	40 (1.9)
Contusion	13 (1.3)	4 (2.6)	16 (1.8)	15 (1.7)	2 (1.3)	37 (1.8)
Pain in extremity	16 (1.6)	7 (4.6)	9 (1.0)	19 (2.1)	1 (0.6)	36 (1.7)
Muscle spasms	13 (1.3)	6 (3.9)	13 (1.5)	10 (1.1)	6 (3.9)	35 (1.7)
Abnormal dreams	3 (0.3)	5 (3.3)	13 (1.5)	9 (1.0)	8 (5.2)	35 (1.7)
Weight increased	20 (2.0)	2 (1.3)	17 (2.0)	14 (1.6)	1 (0.6)	34 (1.6)
Paraesthesia	4 (0.4)	2 (1.3)	14 (1.6)	13 (1.5)	4 (2.6)	33 (1.6)
Decreased appetite	13 (1.3)	2 (1.3)	12 (1.4)	14 (1.6)	4 (2.6)	32 (1.5)
Oedema peripheral	21 (1.2)	2 (1.3)	16 (1.8)	9 (1.0)	4 (2.6)	31 (1.5)
Rash	9 (0.9)	3 (2.0)	11 (1.3)	13 (1.5)	2 (1.3)	29 (1.4)

(Table continues)

Preferred Term[a]	Placebo N=1010 n (%)	Istradefylline 10 mg/day N=153 n (%)	Istradefylline 20 mg/day N=869 n (%)	Istradefylline 40 mg/day N=896 n (%)	Istradefylline 60 mg/day N=155 n (%)	Istradefylline Total N=2073 n (%)
Vomiting	9 (0.9)	1 (0.7)	12 (1.4)	15 (1.7)	1 (0.6)	29 (1.4)
Freezing phenomenon	13 (1.3)	4 (2.6)	9 (1.0)	12 (1.3)	3 (1.9)	28 (1.4)
Orthostatic hypotension	11 (1.1)	2 (1.3)	19 (2.2)	6 (0.7)	0	27 (1.3)
Sleep disorder	6 (0.6)	1 (0.7)	14 (1.6)	10 (1.1)	2 (1.3)	27 (1.3)
Depression	15 (1.5)	0	13 (1.5)	9 (1.0)	4 (2.6)	26 (1.3)
Cough	7 (0.7)	1 (0.7)	9 (1.0)	13 (1.5)	2 (1.3)	25 (1.2)
Dyspepsia	13 (1.3)	6 (3.9)	9 (1.0)	8 (0.9)	1 (0.6)	24 (1.2)
Balance disorder	10 (1.0)	4 (2.6)	7 (0.8)	8 (0.9)	4 (2.6)	23 (1.1)
Lipase increased	18 (1.8)	0	11 (1.3)	11 (1.2)	1 (0.6)	23 (1.1)
Hallucination, visual	4 (0.4)	2 (1.3)	7 (0.8)	12 (1.3)	2 (1.3)	23 (1.1)
Vertigo	5 (0.5)	2 (1.3)	8 (0.9)	11 (1.2)	1 (0.6)	22 (1.1)
Blood urea increased	5 (0.5)	1 (0.7)	6 (0.7)	12 (1.3)	2 (1.3)	21 (1.0)
Musculoskeletal pain	11 (1.1)	3 (2.0)	5 (0.6)	13 (1.5)	0	21 (1.0)
Dry mouth	8 (0.8)	0	5 (0.6)	11 (1.2)	4 (2.6)	20 (1.0)
Neck pain	7 (0.7)	4 (2.6)	9 (1.0)	5 (0.6)	2 (1.3)	20 (1.0)
Hypertension	7 (0.7)	0	8 (0.9)	12 (1.3)	0	20 (1.0)

Pool 2 (OL, Long-term Studies)

The most frequently reported TEAEs in the total istradefylline group in Pool 2 were dyskinesia (36.3%), (worsening symptoms of) PD (24.3%), fall (16.2%), insomnia (15.0%), and constipation (15.0%)

Table 106: TEAE Reported for ≥ 10% of Subjects in the Total Istradefylline Group by Preferred Term: Pool 2

	Total Istradefylline (20-60 mg/day) N=2132 n (%)
Subjects with any TEAE	1913 (89.7)
Preferred Term[a]	
Dyskinesia	773 (36.3)
Parkinson's disease[b]	518 (24.3)
Fall	345 (16.2)
Insomnia	319 (15.0)
Constipation	319 (15.0)
Nausea	265 (12.4)
Dizziness	262 (12.3)
Hallucination	229 (10.7)
Back pain	232 (10.9)

Long-term Adverse Effects

Table 107: Subjects with TEAEs Reported for ≥ 10% of Subjects in the Total Istradefylline Group in Pool 2 by Preferred Term and Event Rate with Corresponding Rates for Placebo Subjects in Pool 1

	Pool 1 Placebo N=1010 (PY=229.8)		Pool 2 Total Istradefylline (20-60 mg/day) N=2132 (PY=2853.9)	
Preferred Term[a]	Number (%) of Subjects with TEAEs	Event Rate Per Subject-yrs of Exposure[b]	Number (%) of Subjects with TEAEs	Event Rate Per Subject-yrs of Exposure[b]
Subjects with any TEAE	661 (65.4)	8.46	1913 (89.7)	6.55
Dyskinesia	97 (9.6)	0.44	773 (36.3)	0.46
Parkinson's disease[c]	37 (3.7)	0.17	518 (24.3)	0.29
Fall	50 (5.0)	0.32	345 (16.2)	0.24
Insomnia	42 (4.2)	0.19	319 (15.0)	0.13
Constipation	33 (3.3)	0.15	319 (15.0)	0.14
Nausea	46 (4.6)	0.22	265 (12.4)	0.12
Dizziness	42 (4.2)	0.22	262 (12.3)	0.12
Hallucination	18 (1.8)	0.09	229 (10.7)	0.11
Back pain	29 (2.9)	0.13	232 (10.9)	0.10

In particular, the analysis of TEAEs that occurred with a higher incidence in the istradefylline monotherapy group compared with the placebo group, revealed that headache (4.3% vs. 3.7%), and fatigue (3.0% vs. 0)

Adverse Events of Special Interest

Hallucination and Other Psychotic disorders

The incidence of the adverse event of special interest (AEoSI) term hallucination was 2.5% in the placebo group and 3.3%, 3.1% and 4.6% in the istradefylline 10, 20, and 40 mg/day groups, respectively in Pool 1. The incidence of the AEoSI term hallucination was highest in the istradefylline 60 mg/day group (7.1%). The AEoSI term hallucination led to discontinuation of treatment in 0.1% of subjects in the placebo group and 0.2% and 1.0% of subjects in the istradefylline 20 and 40 mg/day groups, respectively.

Table 108: Subjects with any TEAE Representing Other Psychotic Disorders in Pool 1

		Istradefylline				
Preferred Term	Placebo N=1010 n (%)	10 mg/day N=153 n (%)	20 mg/day N=869 n (%)	40 mg/day N=896 n (%)	60 mg/day N=155 n (%)	Total N=2073 n (%)
Delusion	1 (0.1%)	1 (0.7%)	1 (0.1%)	3 (0.3%)	0	5 (0.2%)
Paranoia	1 (0.1%)	0	1 (0.1%)	2 (0.2%)	1 (0.6%)	4 (0.2%)
Mania	0	0	0	3 (0.3%)	0	3 (0.1%)
Psychotic Disorder	0	0	1 (0.1%)	1 (0.1%)	0	2 (0.1%)
Persecutory Delusion	0	0	0	1 (0.1%)	0	1 (<0.1%)
Psychiatric Symptom	0	0	1 (0.1%)	0	0	1 (<0.1%)

Table 109: Subjects with any TEAE Representing Psychotic Behaviors in Pool 1

Preferred Term	Placebo N=1010 n (%)	Istradefylline 10 mg/day N=153 n (%)	Istradefylline 20 mg/day N=869 n (%)	Istradefylline 40 mg/day N=896 n (%)	Istradefylline 60 mg/day N=155 n (%)	Istradefylline Total N=2073 n (%)
Confusional State	2 (0.2%)	5 (3.3%)	6 (0.7%)	5 (0.6%)	2 (1.3%)	18 (0.9%)
Agitation	0	0	2 (0.2%)	3 (0.3%)	2 (1.3%)	7 (0.3%)
Delirium	0	0	2 (0.2%)	2 (0.2%)	0	4 (0.2%)
Abnormal Behaviour	0	0	3 (0.3%)	0	0	3 (0.1%)
Anger	0	0	0	1 (0.1%)	0	1 (<0.1%)
Disorientation	2 (0.2%)	0	0	1 (0.1%)	0	1 (<0.1%)
Aggression	2 (0.2%)	0	0	0	0	0

Subjects with Serious TEAEs by SOC and PT
- Pool 1

System Organ Class/ Preferred Term [a]	Placebo (N=1010) n(%) of Subjects	Placebo (N=1010) Total AE(n)	Istradefylline 10 mg/day (N=153) n(%) of Subjects	Istradefylline 10 mg/day (N=153) Total AE(n)	Istradefylline 20 mg/day (N=869) n(%) of Subjects	Istradefylline 20 mg/day (N=869) Total AE(n)	Istradefylline 40 mg/day (N=896) n(%) of Subjects	Istradefylline 40 mg/day (N=896) Total AE(n)	Istradefylline 60 mg/day (N=155) n(%) of Subjects	Istradefylline 60 mg/day (N=155) Total AE(n)	Total Istradefylline (N=2073) n(%) of Subjects	Total Istradefylline (N=2073) Total AE(n)
Psychiatric Disorders	1 (0.1%)	1	0	0	3 (0.3%)	5	7 (0.8%)	9	0	0	10 (0.5%)	14
Confusional State	1 (0.1%)	1	0	0	1 (0.1%)	1	2 (0.2%)	2	0	0	3 (0.1%)	3
Delirium	0	0	0	0	1 (0.1%)	1	2 (0.2%)	2	0	0	3 (0.1%)	3
Psychotic Disorder	0	0	0	0	1 (0.1%)	1	1 (0.1%)	1	0	0	2 (0.1%)	2
Depression	0	0	0	0	0	0	1 (0.1%)	1	0	0	1 (<0.1%)	1
Disorientation	0	0	0	0	1 (0.1%)	1	0	0	0	0	1 (<0.1%)	1
Hallucination	0	0	0	0	0	0	1 (0.1%)	1	0	0	1 (<0.1%)	1
Persecutory Delusion	0	0	0	0	0	0	1 (0.1%)	1	0	0	1 (<0.1%)	1
Psychiatric Symptom	0	0	0	0	1 (0.1%)	1	0	0	0	0	1 (<0.1%)	1
Suicide Attempt	0	0	0	0	0	0	1 (0.1%)	1	0	0	1 (<0.1%)	1

Imbalances for psychiatric disorders including Anxiety and Psychotic disorders were observed. In addition, the TEAEs mania (3 [0.3%] subjects receiving 40 mg/day of istradefylline), agitation (20 mg 2 [0.2%] and 40 mg 3 [0.3%]), delirium (20 mg and 40 mg 2 each [0.2%]), and abnormal behaviour (20 mg 3 [0.3%]) were reported only in the proposed doses of istradefylline versus no cases in the placebo group.

Sleep Disorders

Table 110: Subjects with any TEAE Representing Sleep Disorders in Pool 1

Preferred Term	Placebo N=1010 n (%)	Istradefylline 10 mg/day N=153 n (%)	Istradefylline 20 mg/day N=869 n (%)	Istradefylline 40 mg/day N=896 n (%)	Istradefylline 60 mg/day N=155 n (%)	Istradefylline Total N=2073 n (%)
Insomnia	42 (4.2)	10 (6.5)	31 (3.6)	48 (5.4)	5 (3.2)	94 (4.5)
Initial Insomnia	0	0	1 (0.1)	1 (0.1)	0	2 (0.1)
Middle Insomnia	1 (0.1)	0	3 (0.3)	1 (0.1)	0	4 (0.2)
Terminal Insomnia	1 (0.1)	0	0	3 (0.3)	0	3 (0.1)
Somnolence	32 (3.2)	5 (3.3)	28 (3.2)	17 (1.9)	4 (2.6)	54 (2.6)
Abnormal Dreams	3 (0.3)	5 (3.3)	13 (1.5)	9 (1.0)	8 (5.2)	35 (1.7)
Sleep Disorder	6 (0.6)	1 (0.7)	14 (1.6)	10 (1.1)	2 (1.3)	27 (1.3)
Irregular Sleep Phase	1 (0.1)	2 (1.3)	4 (0.5)	3 (0.3)	1 (0.6)	10 (0.5)
Nightmare	4 (0.4)	0	5 (0.6)	3 (0.3)	0	8 (0.4)
Rapid Eye Movements Sleep Abnormal	0	0	1 (0.1)	5 (0.6)	0	6 (0.3)
Restless Legs Syndrome	2 (0.2)	0	1 (0.1)	3 (0.3)	0	4 (0.2)
Poor Quality Sleep	0	0	0	0	2 (1.3)	2 (0.1)
Sleep Terror	0	0	1 (0.1)	0	1 (0.6)	2 (0.1)
Somnambulism	0	0	1 (0.1)	0	1 (0.6)	2 (0.1)
Hypersomnia	0	0	1 (0.1)	0	0	1 (<0.1)
Sleep Attacks	0	0	1 (0.1)	0	0	1 (<0.1)
Sleep Talking	0	0	1 (0.1)	0	0	1 (<0.1)
Sudden onset of sleep	0	0	0	1 (0.1)	0	1 (<0.1)

Sleep disorders had a slightly higher frequency in the istradefylline group in Pool 1 such as abnormal dreams (All 1.5%, 20 mg 1.4%, 40 mg 0.9% vs. placebo 0.3%), or in patients treated with the highest proposed dose of 40 mg, such as insomnia (4.1% vs. 3.2% in the placebo group). In addition, sleep disorders were one of the most commonly reported AEs in Pool 2.

Suicidality

Table 111: Suicidality: Pools 1 and 2

AEoSI Preferred Term[a]	TEAEs in Pool 1[b] Placebo N=1010 n (%)	TEAEs in Pool 1[b] Istradefylline[c] 20 mg/day N=869 n (%)	TEAEs in Pool 1[b] Istradefylline[c] 40 mg/day N=896 n (%)	TEAEs in Pool 2[d] Total Istradefylline (20-60 mg/day) N=2132 n (%)
Suicidality	0	1 (0.1)	1 (0.1)	13 (0.6)
Depression suicidal	0	0	0	1 (<0.1)
Suicidal behavior	0	0	0	1 (<0.1)
Suicidal ideation	0	1 (0.1)	0	9 (0.4)
Suicide attempt	0	0	1 (0.1)	2 (0.1)

Impulse Control Disorder

The incidence of the AEoSI term ICD was 0% in the placebo group and 0.7%, 0.8%, 0.6%, and 0% in the istradefylline 10, 20, 40 and 60 mg/day groups, respectively in Pool 1 The AEoSI term ICD led to discontinuation of treatment in 1 subject in the istradefylline 20 mg/day group (0.1%)

A detailed review of ICDs in Pool 1 data revealed that 11 out of 13 subjects who experienced ICDs were concomitantly taking DAs for several months prior to istradefylline initiation. In the majority of the subjects who were concomitantly taking DAs, no action was taken regarding istradefylline treatment following development of ICDs (9 out of 11 subjects); however, the DA dose was decreased or discontinued in 6 out of 9 subjects (events were reported as resolved in 5 subjects) and DAs was continued in 3 out of 9 subjects (events reported as resolved in 2 subjects). In the additional 2 subjects who were taking istradefylline and DAs concomitantly, the events resolved following dose reduction and discontinuation of both istradefylline and DA.

Table 112: Subjects with AEoSI: Impulse Control Disorder by PT - Pool 2

	Total Istradefylline (N=2132)	
Preferred Term [a]	n(%) of Subjects	Total AE(n)
Subjects with any TEAE	67 (3.1%)	81
Obsessive-Compulsive Disorder	33 (1.5%)	35
Libido Increased	11 (0.5%)	14
Gambling Disorder	10 (0.5%)	10
Disturbance In Sexual Arousal	6 (0.3%)	8
Impulse-Control Disorder	4 (0.2%)	6
Compulsions	2 (0.1%)	2
Hypersexuality	2 (0.1%)	2
Impulsive Behaviour	2 (0.1%)	2
Compulsive Sexual Behaviour	1 (<0.1%)	1
Excessive Sexual Fantasies	1 (<0.1%)	1

Cardiac Safety Assessment

The incidence of TEAEs in the cardiac disorders SOC was higher in the placebo group (3.5%) and istradefylline 60 mg/day group (5.2%) than in the istradefylline 10 mg/day (2.0%), istradefylline 20 mg/day (3.0%) or istradefylline 40 mg/day (2.8%) groups (see the table below). The exposure-adjusted event rates from long-term treatment (Pool 2) do not suggest a higher risk of cardiac events with longer istradefylline treatment compared with placebo.

Table 113: Overview of TEAEs in the Cardiac Disorders SOC in Pool 1 and Pool 2

	Pool 1										**Pool 2**	
			Istradefylline								**Istradefylline**	
	Placebo N=1010 (PY=229.8)		**10 mg/day N=153 (PY=33.5)**		**20 mg/day N=869 (PY=190.1)**		**40 mg/day N=896 (PY=207.4)**		**60 mg/day N=155 (PY=31.9)**		**20-60 mg/day N=2132 (PY=2853.9)**	
Category	N (%)	EAIR	N (%)	EAIR	N (%)	EAIR	N (%)	EAIR	N (%)	EAIR	N (%)	EAIR
TEAEs	35 (3.5)	0.18	3 (2.0)	0.09	26 (3.0)	0.15	25 (2.8)	0.16	8 (5.2)	0.25	172 (8.1)	0.09
Treatment-related TEAEs	18 (1.8)	-	1 (0.7)	-	17 (2.0)	-	15 (1.7)	-	3 (1.9)	-	84 (3.9)	-
SAEs	8 (0.8)	-	3 (2.0)	-	3 (0.3)	-	8 (0.9)	-	1 (0.6)	-	51 (2.4)	-
Treatment-related SAEs	2 (0.2)	-	1 (0.7)	-	1 (0.1)	-	5 (0.6)	-	0	-	18 (0.8)	-

Table 114: TEAEs in the Cardiac Disorders SOC Reported for ≥ 2 Subjects in Pool 1

System Organ Class Preferred Term[a]	Placebo N=1010 n (%)	Istradefylline 10 mg/day N=153 n (%)	Istradefylline 20 mg/day N=869 n (%)	Istradefylline 40 mg/day N=896 n (%)	Istradefylline 60 mg/day N=155 n (%)
Cardiac Disorders SOC	35 (3.5)	3 (2.0)	26 (3.0)	25 (2.8)	8 (5.2)
Angina pectoris	4 (0.4)	0	0	3 (0.3)	0
Atrial fibrillation	2 (0.2)	0	2 (0.2)	3 (0.3)	0
Atrioventricular block first degree	2 (0.2)	0	0	0	1 (0.6)
Bradycardia	5 (0.5)	0	1 (0.1)	2 (0.2)	0
Bundle branch block right	2 (0.2)	0	1 (0.1)	2 (0.2)	0
Cardiac failure congestive	0	0	2 (0.2)	2 (0.2)	0
Coronary artery disease	3 (0.3)	0	0	0	0
Left ventricular hypertrophy	0	0	0	2 (0.2)	0
Myocardial infarction	1 (0.1)	0	0	2 (0.2)	0
Palpitations	4 (0.4)	0	7 (0.8)	3 (0.3)	2 (1.3)
Supraventricular extrasystoles	2 (0.2)	1 (0.7)	2 (0.2)	1 (0.1)	0
Tachycardia	1 (0.1)	0	2 (0.2)	0	0
Tricuspid valve incompetence	0	0	0	2 (0.2)	0
Ventricular extrasystoles	5 (0.5)	0	2 (0.2)	2 (0.2)	2 (1.3)
Wolff-Parkinson-White syndrome	2 (0.2)	0	0	0	0

Twenty-four subjects in Pools 1, 2 and 3 who received istradefylline were reported to have treatment-related SAEs in the cardiac disorders SOC, including 3 subjects who had cardiac events with an outcome of death. All 24 subjects had significant cardiac medical history and/or concurrent medical conditions. a. Although there are not differences in the incidence of most TEAEs in the SOC Cardiac disorders, there are 3 SAEs of cardiac failure congestive (1 for 20 mg dose and 2 for 40 mg dose) and 2 SAEs of myocardial infarction (2 for 40 mg dose) in the istradefylline treated patients versus none in the placebo group.

Of the estimated 63,500 patients treated with istradefylline in the post-marketing setting as of 31 May 2019, a total of 32 (2% of all AEs) cardiac events were reported, including 23 serious events and 9 non-serious events. No safety signal relating to cardiac safety was identified from the post-marketing evaluation.

In particular, in Pool 4a, the TEAEs potentially representing lung inflammation included upper respiratory tract infections reported in 5.5% (9/164) of subjects in the istradefylline 40 mg/day group and 3.7% (3/82) in the placebo group; bronchitis reported in 1.2% (1/82) of subjects in placebo group (none in istradefylline 40 mg/day group); dyspnoea reported in 1.2% (1/82) of subjects in placebo group and 0.6% (1/164) of subjects in istradefylline 40 mg/day group; and pleural effusion reported in 0.6% (1/164) of subjects in istradefylline 40 mg/day group (none in placebo group). The higher difference from placebo of upper respiratory tract infections and viral upper respiratory tract infections in the istradefylline monotherapy group in Pool 4a compared with Pool 1, was observed in the absence of apparent plausible reasons (e.g. substantial similar demographics and baseline disease characteristics

between the two treatment groups, no higher incidence of dysphagia or haematology alterations in the istradefylline patients).

As regard to orthostatic hypotension, in Pool 1, orthostatic hypotension-related AEs were reported in the 9.4% and 7.4% of patients treated with istradefylline 20 mg/day and 40 mg/day, respectively, versus 6.2% of placebo patients. The most frequently TEAE related to orthostatic hypotension was dizziness (20 mg 5.1% and 40 mg 5.9% vs. 4.2% in the placebo group), but also the specific TEAE orthostatic hypotension was reported with a higher incidence in the istradefylline group, doubled compared to placebo (2.2% and 0.7% vs. 1.1%).

Brain vascular mineralization

Pre-clinical data

As part of the istradefylline development program, two year carcinogenicity studies with istradefylline were performed in rats and mice and long term (6 and 12 months) toxicology studies with istradefylline were conducted in dogs.

Histopathological examination of the rodent brains, however, revealed foci of mineralization which were usually evident in the walls of arterioles and small arteries primarily located in the basal ganglia and, more rarely, in other parts of the brain. The foci were observed in both control and istradefylline treated mice, with prevalence increasing as a function of age and unrelated to the dose of istradefylline. In rats, the foci of mineralization were observed rarely in control animals but increased with dose and exposure duration in istradefylline treated rats. The foci of mineralisations in rats and mice were not associated with evidence of neuronal damage, inflammation, or gliosis.

In the dog studies, no foci of vascular mineralization or any other abnormalities including neuronal damage, inflammation or gliosis were observed in the brain with or without exposure to istradefylline.

The Sponsor's position is that the finding in rats is a species-specific effect, posing a low potential risk in terms of human safety.

The istradefylline plasma concentrations in rats were at least four times higher than those anticipated in humans. Accordingly, the potential safety risk in humans was considered to be low.

Foci of mineralization of the brain in humans

There is a rare neurological condition which provides a potential model of what might be identified as "primary" mineralization in the basal ganglia and other parts of the brain. Primary familial brain calcification (PFBC) is a name for this condition that has been identified as familial idiopathic basal ganglia calcification, striatopallidodentate calcinosis, non-arteriosclerotic vascular mineralization of the basal ganglia, and Fahr's disease among many others (Casanova, 2003; Manyam, 2005; Oliveira, 2011; Ramos, 2017; Betsholtz, 2014). This condition is characterized by abnormal mineral deposits in the basal ganglia, dentate nucleus, thalamus, and cerebral white matter.

Some patients with imaging-confirmed PFBC present clinically with various combinations of neurological and psychiatric symptoms. These can include dementia, psychosis, mood swings, loss of motor skills, rigidity, and spastic paralysis. Dystonia, athetosis and chorea have also been observed in some patients. In addition, Parkinsonian features such as tremors and rigidity, masklike facial expression, shuffling walk, and pill-rolling have been reported (Casanova, 2003; Manyam, 2005; Oliveira, 2011; Ramos, 2017).

Istradefylline safety data

The istradefylline safety database, however, is much larger now (nearly 50,000 clinical trial subjects and patients as of 31 Mar 2018) than it was in the 2007 NDA submission. Accordingly, clinical trial experience

with istradefylline was examined for signs and symptoms bearing on the possible development of brain vascular mineralization. Likewise, the postmarketing safety database was reviewed for signals that could be ascribed to vascular mineralization in the brains of istradefylline-treated patients. AES related to movement disorders in the postmarketing safety database were identified within the nervous system disorders SOC. The psychiatric system disorders SOC was also reviewed.

In addition, because clinical trial experience with istradefylline is broader now than in 2007, meta-analyses and reviews have also been published (Muller, 2015; Chen, 2013; Tao, 2015; Sako, 2017) that provide another perspective on the clinical safety of istradefylline.

More than 4300 subjects received istradefylline in completed Phase 1, 2, and 3 studies. There were 3501 subjects who took at least 1 dose of istradefylline in a Phase 2 or 3 study (Table 99).

Neither imaging studies nor autopsies were required elements in the clinical studies. Without brain imaging or autopsy-derived information indicating the presence or absence of brain vascular mineralization, it is not possible to state definitively that no brain vascular mineralization occurred. It is noteworthy, however, that no adverse events of 'mineralization' or 'brain mineralization' were reported.

As noted previously, it is difficult to identify adverse events that distinguish possible brain vascular mineralization from the signs and symptoms of PD and PD medications. It might be expected, however, that motor function would be impaired and certain adverse events (related to 'worsening of Parkinson's disease' and 'cognition and memory') would be increased compared to placebo if brain vascular mineralization serious enough to be symptomatic had developed. In the ISS, pooled data from 8 placebo-controlled Phase 2/3 studies of 12 or 16 weeks duration (Pool 1: Studies 6002-US-005, 6002-US-006, 6002-US-013, 6002-US-018, 6002-EU-007, 6002-0608, 6002-009, and 6002-014) provide useful information to address this issue.

Pool 1 includes a placebo group for comparison with istradefylline-treated groups. There were 2073 subjects who received istradefylline and 1010 who received placebo. Subjects in the studies had idiopathic PD and had motor response complications with L/C or levodopa/benserazide therapy, i.e., the patient population targeted in the proposed indication. It is important to note that all subjects, including subjects in the placebo group, were receiving levodopa, in combination with a peripheral dopa-decarboxylase inhibitor, and possibly other additional standard anti-Parkinson medication. Most of the subjects in Pool 1 who received istradefylline were treated with the proposed dosing regimen of 20 or 40 mg/day (1765 of 2073 subjects).

Pool 1 includes results of UPDRS Part III (ON) evaluations which provide a means of assessing the effect of istradefylline on motor function. If brain vascular mineralization had developed, it might be expected that UPDRS Part III (ON) scores would increase, reflecting impairment of motor performance. In Pool 1, the reduction from baseline (indicating improvement) in UPDRS Part III (ON) scores with istradefylline was greater than with placebo treatment in 7 of 8 studies, although not statistically separated from placebo. In pooled results from all 8 studies in Pool 1, UPDRS Part III (ON) scores in the istradefylline treatment groups showed modest improvement, compared with placebo. That is, the UPDRS motor scores did not worsen, which would probably not be the case had brain vascular mineralization developed.

The AE of falls represents another potential sign that could be associated with brain vascular mineralization since falls may also indicate impaired motor function. In the ISS, the incidence of falls (including TEAEs related to falls, fractures, and injury) in Pool 1 was similar in the placebo group and in the istradefylline 20 mg/day and 40 mg/day groups (7.6%, 7.9%, and 7.7%, respectively).

Symptomatic brain vascular mineralization might be expected to adversely affect memory and cognition. In Pool 1, istradefylline treatment did not appear to be associated with greater impairment of memory

and cognition, compared to placebo. The incidence of memory or cognition impairment was 0.8% in the placebo group and 1.4% and 0.7% in the istradefylline 20 mg/day and 40 mg/day groups, respectively.

Considering the overlap in signs and symptoms of brain vascular mineralization with those of PD, the adverse event term, PD (which represents worsening symptoms of PD), is another potentially relevant adverse event appearing in the clinical trial database. In Pool 1, the frequency of the adverse event PD was similar in the placebo group and in the 20 mg/day and 40 mg/day groups (3.7%, 3.6%, and 2.8%, respectively).

In summary, the safety and efficacy data from the double-blind, randomized, placebo-controlled Phase 2b/3 clinical trials did not show worsening of motor function based on UPDRS Part III (ON) scores, or more frequent adverse events of falls, memory or cognition impairment, or (worsening symptoms of) PD with istradefylline treatment, compared with placebo. The Sponsor concludes that this method of searching the clinical trial database produced no evidence suggesting the development of brain vascular mineralization during 12 or 16 weeks of istradefylline treatment.

Post –marketing data

There was a low number of movement disorder and psychiatry-related adverse events in the post-marketing safety database of approximately 46500 istradefylline-treated patients. Since most of the events were characterized by rapid onset and/or rapid resolution, it is unlikely they would have been secondary to relatively gradual development and persistence of vascular mineralization in the brain. In addition, the events occurred on a background of variable disease burden, co-morbidities, and concomitant medications that represent alternative (or, at least, additional) explanations for the adverse events. Since confirmatory imaging studies are lacking, it is not possible to rule out the occurrence of brain vascular mineralization. Nevertheless, the findings from the post-marketing safety database did not indicate the occurrence of istradefylline-associated brain vascular mineralization that was serious enough to be symptomatic.

In summary, there was a low number of movement disorder and psychiatry-related adverse events in the post-marketing safety database of approximately 46500 istradefylline-treated patients. Since most of the events were characterized by rapid onset and/or rapid resolution, it is unlikely they would have been secondary to relatively gradual development and persistence of vascular mineralization in the brain. In addition, the events occurred on a background of variable disease burden, co-morbidities, and concomitant medications that represent

Serious adverse event/deaths/other significant events

Deaths

One subject died in the Phase 1 development program.

Nine subjects died in Pool 1. The most frequently reported TEAEs with an outcome of death were those related with cardio-respiratory events.

Thirty-two subjects died in Pool 2. All subjects in Pool 2 were treated with istradefylline. The most frequently reported TEAEs with an outcome of death were pneumonia (7 subjects [0.3%]), aspiration pneumonia (4 subjects [0.2%]), (worsening symptoms of) PD (3 subjects [0.1%]), and cardiac arrest (3 subjects [0.1%]).

Other Serious Adverse Events

Pool 1 (DB, Placebo-controlled, Fixed-dose Studies)

The incidence of serious TEAEs was 4.2% in the total istradefylline group and 3.1% in the placebo group in these 12- or 16-week studies, with similar incidences in the istradefylline 20 mg/day (3.9%) and 40 mg/day (4.8%) treatment groups.

The SOCs with ≥ 0.5% incidence of serious TEAEs in the total istradefylline group were cardiac disorders (0.7% istradefylline; 0.8% placebo), infections and infestations (0.7% istradefylline; 0.6% placebo), injury, poisoning and procedural complications (0.7% istradefylline; 0.6% placebo), nervous system disorders (0.5% istradefylline; 0.4% placebo), and psychiatric disorders (0.5% istradefylline; 0.1% placebo).

The most frequently reported serious TEAEs were falls and pneumonia, both of which were reported with similar or a lower incidence in the istradefylline 20 mg/day and 40 mg/day groups compared with the placebo group. Most of these events were not considered related to treatment by the Investigator.

Serious TEAEs of cardiac failure congestive, delirium, and rib fracture occurred more frequently in both the istradefylline 20 mg/day and 40 mg/day groups (1 or 2 subjects, 0.1% or 0.2%, respectively) compared with the placebo group (0%). All serious TEAEs of delirium and rib fracture were considered unrelated to study drug. Cardiac events are discussed in detail below.

Table 115: Serious TEAEs Reported for > 2 Subjects in the Total Istradefylline Group with Corresponding Treatment-related Incidence by Preferred Term: Pool 1

	Serious TEAEs						Related Serious TEAEs
		Istradefylline					Istradefylline
	Placebo N=1010 n (%)	10 mg/day N=153 n (%)	20 mg/day N=869 n (%)	40 mg/day N=896 n (%)	60 mg/day N=155 n (%)	Total N=2073 n (%)	Total N=2073 n (%)
Subjects with any serious TEAE	31 (3.1)	6 (3.9)	34 (3.9)	43 (4.8)	4 (2.6)	87 (4.2)	30 (1.4)
Preferred Term[a]							
Fall	2 (0.2)	1 (0.7)	2 (0.2)	3 (0.3)	1 (0.6)	7 (0.3)	1 (<0.1)
Pneumonia	4 (0.4)	1 (0.7)	1 (0.1)	2 (0.2)	1 (0.6)	5 (0.2)	1 (<0.1)
Cardiac failure congestive	0	0	1 (0.1)	2 (0.2)	0	3 (0.1)	3 (0.1)
Confusional state	1 (0.1)	0	1 (0.1)	2 (0.2)	0	3 (0.1)	1 (<0.1)
Delirium	0	0	1 (0.1)	2 (0.2)	0	3 (0.1)	0
Hip fracture	0	0	2 (0.2)	0	1 (0.6)	3 (0.1)	0
Parkinson's disease[b]	1 (0.1)	1 (0.7)	1 (0.1)	1 (0.1)	0	3 (0.1)	1 (<0.1)
Rib fracture	0	0	1 (0.1)	2 (0.2)	0	3 (0.1)	0

Pool 2 (OL, Long-term Studies)

Serious TEAEs were reported for 22.6% of subjects in the total istradefylline group.

The SOCs with ≥ 2% incidence of serious TEAEs in the total istradefylline group were nervous system disorders (6.8%), injury, poisoning and procedural complications (4.5%), infections and infestations (3.6%), musculoskeletal and connective tissue disorders (3.4%), psychiatric disorders (3.0%), gastrointestinal disorders (2.5%), and cardiac disorders (2.4%),

The most frequently reported serious TEAEs in the total istradefylline group were (worsening symptoms of) PD (3.2%), pneumonia (1.4%), and fall (1.3%), all of which are not unanticipated in a population of elderly subjects (median age: 64.0 years) with PD (Woodford, 2005). All other serious TEAEs were reported for < 1.0% of subjects in the total istradefylline group.

Syncope was reported as a serious TEAE for 16 subjects (0.8%) in Pool 2. Most of these subjects had concurrent medical conditions, such as, cardiac disease and/or hypotension.

In Pool 2, confusional state was reported for 4.4% of subjects in the total istradefylline group. the incidence of serious confusional state was 0.8% In Pool 2, the incidence of psychotic disorder and delusion was 0.9% each

Table 116: Serious TEAEs in ≥ 0.5% of Subjects and Additional Treatment-related Serious TEAEs (Reported in ≥ 3 Subjects) in the Total Istradefylline Group by Preferred Term: Pool 2

	Total Istradefylline (20-60 mg/day) N=2132 n (%)	
	All Serious TEAEs	Treatment-related Serious TEAEs
Subjects with any serious TEAE	482 (22.6)	127 (6.0)
Preferred Term[a]		
Serious TEAEs in ≥ 0.5% of Subjects		
Parkinson's disease[b]	69 (3.2)	6 (0.3)
Pneumonia	29 (1.4)	3 (0.1)
Fall	28 (1.3)	3 (0.1)
Osteoarthritis	18 (0.8)	1 (<0.1)
Confusional state	17 (0.8)	9 (0.4)
Hip fracture	16 (0.8)	1 (<0.1)
Syncope	16 (0.8)	12 (0.6)
Hallucination	13 (0.6)	12 (0.6)
Psychotic disorder	11 (0.5)	8 (0.4)
Spinal column stenosis	11 (0.5)	0
Chest pain	10 (0.5)	1 (<0.1)
Myocardial infarction	10 (0.5)	5 (0.2)
Coronary artery disease	10 (0.5)	2 (0.1)
		Treatment-related Serious TEAEs in ≥ 3 Subjects
Dyskinesia	6 (0.3)	4 (0.2)
Transient ischaemic attack	6 (0.3)	3 (0.1)
Delusion	4 (0.2)	4 (0.2)
Agitation	4 (0.2)	3 (0.1)
Anxiety	4 (0.2)	3 (0.1)

Laboratory findings

Haematology

Pool 1 (DB, Placebo-controlled, Fixed-dose Studies)

There were no clinically meaningful mean changes from Baseline to Endpoint in the haematology analytes in Pool 1.

Shifts from normal-to-low or normal-to-high were observed in < 10% of subjects in any dose group for all of the haematology analytes in Pool 1. The incidences of subjects with shifts were similar in the placebo and in the istradefylline 20 mg/day and 40 mg/day groups, i.e., ≤ 5% difference between placebo and istradefylline 20 mg/day and 40 mg/day groups. The incidence of subjects with normal-to-low or normal-to-high shifts at each visit during treatment was similar in the placebo and in the 20 mg/day and 40 mg/day istradefylline groups in Pool 1.

Less than 5% of subjects in any dose group had at least 1 PCS haematology value in Pool 1. The incidences of subjects with PCS haematology values were similar in the placebo and istradefylline 20 mg/day and 40 mg/day groups.

In particular, the incidences of subjects with neutrophil counts below the lower limit the normal range (≤ 1.0 LLN) at any visit was lower in istradefylline-treated groups than in the placebo group. The incidences of subjects with neutrophil counts ≤ 0.5 x LLN was zero or < 1.0% in all treatment groups, including placebo

Table 117: Subjects with Treatment-emergent PCS Haematology Values at Any Visit During Treatment: Pool 1

		Istradefylline				
PCS Laboratory Analyte	Placebo N=1010 n (%)	10 mg/day N=153 n (%)	20 mg/day N=869 n (%)	40 mg/day N=896 n (%)	60 mg/day N=155 n (%)	Total N=2073 n (%)
Erythrocytes ≤ 0.8 × LLN	7 (<1)	0	8 (<1)	8 (<1)	0	16 (<1)
Erythrocytes ≥ 1.2 × ULN	1 (<1)	0	0	1 (<1)	0	1 (<1)
Hematocrit ≤ 0.8 × LLN	6 (<1)	0	9 (1)	5 (<1)	0	14 (<1)
Hematocrit ≥ 1.2 × ULN	0	0	0	0	0	0
Hemoglobin ≤ 0.8 × LLN	9 (<1)	0	15 (2)	7 (<1)	0	22 (1)
Hemoglobin ≥ 1.2 × ULN	0	0	0	0	0	0
Leukocytes ≤ 0.8 × LLN	32 (3)	2 (1)	21 (2)	17 (2)	1 (<1)	41 (2)
Leukocytes ≥ 1.2 × ULN	13 (1)	1 (<1)	11 (1)	18 (2)	1 (<1)	31 (2)
Neutrophils ≤ 1.0 × LLN	46 (5)	4 (3)	28 (3)	26 (3)	3 (2)	61 (3)
Neutrophils ≤ 0.5 × LLN	1 (<1)	0	2 (<1)	2 (<1)	0	4 (<1)
Neutrophils ≥ 1.5 × ULN	7 (<1)	0	4 (<1)	6 (<1)	1 (<1)	11 (<1)
Eosinophils ≥ 2.0 × ULN	0	0	1 (<1)	2 (<1)	1 (<1)	4 (<1)
Platelets ≤ 0.7 × LLN	6 (<1)	0	5 (<1)	5 (<1)	0	10 (<1)
Platelets ≥ 1.3 × ULN	3 (<1)	0	3 (<1)	3 (<1)	0	6 (<1)

Pool 2 (Open-label, Long-term Studies)

Mean changes from baseline to endpoint for the haematology analytes in the long-term studies (Pool 2) were greater than in Pool 1. Nevertheless, the mean changes from baseline to endpoint remained small and there were no clinically meaningful changes in any of the mean values for haematology analytes from Baseline to Endpoint.

Greater percentages of subjects had shifts from normal-to-low or normal-to-high in Pool 2 compared with the Pool 1. Nevertheless, shifts from normal-to-low or normal-to-high were observed in < 20% of subjects for all of the haematology analytes.

Greater percentages of subjects had PCS haematology values in Pool 2 compared with Pool 1; however, ≤ 5% of subjects in the total istradefylline group in Pool 2 had at least 1 PCS haematology value.

Table 118: Subjects with Treatment-emergent PCS Haematology Values at Any Visit During Treatment: Pool 2

PCS Laboratory Analyte	Total Istradefylline (20-60 mg/day) N=2132 n (%)
Erythrocytes ≤ 0.8 × LLN	36 (2)
Erythrocytes ≥ 1.2 × ULN	3 (<1)
Hematocrit ≤ 0.8 × LLN	27 (1)
Hematocrit ≥ 1.2 × ULN	1 (<1)
Hemoglobin ≤ 0.8 × LLN	52 (2)
Hemoglobin ≥ 1.2 × ULN	1 (<1)
Leukocytes ≤ 0.8 × LLN	56 (3)
Leukocytes ≥ 1.2 × ULN	70 (3)
Neutrophils ≤ 1.0 × LLN	114 (5)
Neutrophils ≤ 0.5 × LLN	6 (<1)
Neutrophils ≥ 1.5 × ULN	42 (2)
Eosinophils ≥ 2.0 × ULN	11 (<1)
Platelets ≤ 0.7 × LLN	17 (<1)
Platelets ≥ 1.3 × ULN	23 (1)

Chemistry

Pool 1 (DB, Placebo-controlled, Fixed-dose Studies)

There were no clinically meaningful mean changes from Baseline to Endpoint in any of the blood chemistry analytes in Pool 1.

Shifts from normal-to-low were reported in 12% and 13% of subjects in the 20 and 40 mg/day istradefylline groups, respectively, for cholesterol, compared with 13% of subjects receiving placebo. Shifts from normal-to-high were reported in ≥ 10% of the subjects in the 20 and/or 40 mg/day istradefylline groups for urea nitrogen, lactate dehydrogenase, glucose, lipase, triglycerides, and creatine kinase (CK)

The incidences of subjects with shifts were similar in the placebo group and the 20 and 40 mg/day istradefylline groups.

Few subjects (≤ 2%) in any dose group had a PCS blood chemistry value in Pool 1 with the exception of glucose ≥ 1.5 x ULN, lipase ≥ 1.5 x ULN, amylase ≥ 1.5 × ULN, cholesterol ≥ 1.25 x ULN, triglycerides ≥ 1.6 x ULN, and CK ≥ 2 x ULN. For these 6 analytes, the incidence of PCS values was ≤ 10% for istradefylline-treated subjects, with ≤ 3% greater incidence in these 2 istradefylline dose groups (20 and 40 mg/day) compared with placebo. The incidence of amylase and lipase both simultaneously increased ≥ 1.5 × ULN was reported by ≤ 1.0% of subjects in any dose group.

Table 119: Subjects with Treatment-emergent PCS Blood Chemistry Values at Any Visit During Treatment: Pool 1

PCS Laboratory Analyte	Placebo N=1010 n (%)	Istradefylline 10 mg/day N=153 n (%)	Istradefylline 20 mg/day N=869 n (%)	Istradefylline 40 mg/day N=896 n (%)	Istradefylline 60 mg/day N=155 n (%)	Istradefylline Total N=2073 n (%)
Creatinine ≥ 1.5 × ULN	0	0	2 (<1)	1 (<1)	1 (<1)	4 (<1)
Urea nitrogen ≥ 1.5 × ULN	19 (2)	0	18 (2)	17 (2)	2 (1)	37 (2)
Potassium ≤ 0.85 × ULN	0	0	0	1 (<1)	0	1 (<1)
Potassium ≥ 1.15 × ULN	1 (<1)	0	0	3 (<1)	0	3 (<1)
Sodium ≤ 0.95 × LLN	3 (<1)	0	2 (<1)	2 (<1)	0	4 (<1)
Sodium ≥ 1.05 × ULN	4 (<1)	1 (<1)	2 (<1)	0	1 (<1)	4 (<1)
Chloride ≤ 0.95 × ULN	4 (<1)	0	1 (<1)	2 (<1)	0	3 (<1)
Chloride ≥ 1.05 × ULN	4 (<1)	1 (<1)	3 (<1)	2 (<1)	0	6 (<1)
AST ≥ 3 × ULN	0	1 (<1)	3 (<1)	5 (<1)	1 (<1)	10 (<1)
AST ≥ 5 × ULN	0	1 (<1)	2 (<1)	1 (<1)	0	4 (<1)
AST ≥ 10 × ULN	0	0	1 (<1)	0	0	1 (<1)
ALT ≥ 3 × ULN	2 (<1)	0	3 (<1)	4 (<1)	0	7 (<1)
ALT ≥ 5 × ULN	0	0	1 (<1)	3 (<1)	0	4 (<1)
ALT ≥ 10 × ULN	0	0	0	0	0	0
Alkaline phosphatase ≥ 3 × ULN	3 (<1)	0	1 (<1)	1 (<1)	0	2 (<1)
LDH ≥ 3 × ULN	0	1 (<1)	1 (<1)	1 (<1)	0	3 (<1)
Calcium ≤ 0.9 × LLN	2 (<1)	0	0	0	0	0
Calcium ≥ 1.1 × ULN	0	0	1 (<1)	0	0	1 (<1)
Bilirubin ≥ 1.6 × ULN	9 (<1)	3 (2)	11 (1)	17 (2)	1 (<1)	32 (2)
Glucose ≤ 0.75 × LLN	8 (<1)	0	3 (<1)	8 (<1)	0	11 (<1)
Glucose ≥ 1.5 × ULN	50 (5)	8 (5)	32 (4)	49 (5)	5 (3)	94 (5)
Urate ≥ 1.2 × ULN	7 (1)	1 (<1)	7 (2)	12 (2)	0	20 (2)
Amylase ≥ 1.5 × ULN	22 (2)	3 (2)	22 (3)	8 (<1)	0	33 (2)
Lipase ≥ 1.5 × ULN	45 (4)	1 (<1)	39 (5)	34 (4)	3 (2)	77 (4)
Trypsin-like immunoreactive ≥ 3.0 × ULN	5 (<1)	1 (<1)	3 (<1)	0	0	4 (<1)
Amylase and lipase both simultaneously ≥ 1.5 × ULN	11 (1)	1 (<1)	6 (<1)	3 (<1)	0	10 (<1)
Amylase and lipase both ≥ 1.5 × ULN and trypsin ≥ 3.0 × ULN simultaneously	2 (< 0.1)	0	0	0	0	0
Albumin ≤ 0.7 × LLN	1 (<1)	0	1 (<1)	0	0	1 (<1)
Albumin ≥ 1.3 × ULN	0	0	0	0	0	0
Cholesterol ≥ 1.25 × ULN	18 (2)	0	19 (2)	23 (3)	0	42 (2)

(Table continues)

PCS Laboratory Analyte	Placebo N=1010 n (%)	Istradefylline 10 mg/day N=153 n (%)	Istradefylline 20 mg/day N=869 n (%)	Istradefylline 40 mg/day N=896 n (%)	Istradefylline 60 mg/day N=155 n (%)	Istradefylline Total N=2073 n (%)
Triglycerides ≥ 1.6 × ULN	57 (6)	9 (6)	53 (6)	60 (7)	6 (4)	128 (6)
Creatine kinase ≥ 2 × ULN	74 (7)	10 (7)	52 (6)	59 (7)	16 (10)	137 (7)

Increased Liver Enzymes

The incidence of AEoSI term increased liver enzymes was 2.0% in the placebo group and 1.3%, 1.2%, 2.5%, and 2.6% in the istradefylline 10, 20, 40, and 60 mg/day groups, respectively. The AEoSI term increased liver enzymes was reported as an SAE for 0.1% of subjects in both the istradefylline 20 and 40 mg/day groups. The AEoSI term increased liver enzymes led to discontinuation of treatment in subjects in the istradefylline 20 (0.1%), 40 (0.4%), and 60 mg/day (0.6%) groups, respectively, compared with none in the placebo group.

In addition to AEoSI of increased liver enzymes, the clinical database for Pools 1 and 2 was searched for any subject meeting Hy's law criteria.

Vital Signs

Pool 1 (DB, Placebo-controlled, Fixed-dose Studies)

There were no clinically meaningful mean changes from Baseline to Endpoint in vital signs (sitting) in Pool 1

Mean weight loss over the 12- or 16-week duration of the Pool 1 studies was < 0.5 kg in all treatment groups. 5% of subjects in any dose group had at least 1 PCS high or low vital sign (sitting) or change from baseline in body weight ≥ 7% at any time in Pool 1.

Pool 2 (OL, Long-term Studies)

There were no clinically meaningful mean changes from Baseline to Endpoint in vital signs (sitting) in Pool 2. Mean body weight decreased by 1.61 kg from Baseline to Endpoint. This decrease in body weight was greater than that observed for istradefylline 40 mg/day in Pool 1 (-0.43 kg) which can be attributed to the longer treatment duration for subjects in Pool 2 compared to Pool 1.

Electrocardiograms

Pool 1 (DB, Placebo-controlled, Fixed-dose Studies)

There were no clinically meaningful mean changes from Baseline to Endpoint in the ECG parameters in Pool 1.

The incidences of subjects with clinically significant abnormal ECG interpretations (according to the Investigator's interpretation) were < 5% at each post Baseline visit and at study endpoint. The incidences of subjects with clinically significant abnormal ECG interpretations were similar in the placebo and in the istradefylline 20 mg/day and 40 mg/day groups in Pool 1.

Less than 6% of subjects in any istradefylline dose group had at least 1 treatment-emergent PCS ECG value in Pool 1 and <3% of subjects in any dose group had treatment-emergent shifts from normal to PCS in ECG parameters at any time post-baseline.

A slightly higher proportion of subjects with TE-PCS ECG value was observed in the two proposed doses of istradefylline compared with placebo (20 mg 5.6% and 40 mg 4.4% vs. placebo 4.2%), in particular differences in QRS interval (3.5% and 4.1% vs. 2.0%) and QTcF (0.8% and 0.3% vs. 0%) were observed, and a higher proportion of subjects had treatment-emergent shifts in QRS interval in the istradefylline group compared to placebo (20 mg 2.4% and 40 mg 2.5% vs. placebo 1.1%).

Pool 2 (OL, Long-term Studies)

There were no clinically meaningful changes in any of the mean values in the ECG parameters between Baseline and the study Endpoint.

The incidences of subjects with clinically meaningful abnormal ECGs (according to the Investigator interpretation) were < 2% at each post-Baseline visit through Week 52.

Less than 7% of subjects treated with istradefylline had at least 1 treatment-emergent PCS ECG value in Pool 2. Less than 3% of istradefylline-treated subjects had treatment-emergent shifts from normal to PCS in ECG parameters at any time post-baseline.

In Pool 2, findings for the QRS interval were similar to those recorded in Pool 1.

Safety in special populations

Table 120: Overall Summary of TEAEs by Subgroup- Pool 1

	Pool 1			
	Placebo N=1010		Total Istradefylline N=2073	
Subgroup	n/N_1	%	n/N_1	%
Gender				
Female	293/422	69.4	632/842	75.1
Male	368/588	62.6	869/1231	70.6
Age				
< 65 years	362/539	67.2	746/1046	71.3
≥ 65 years	299/471	63.5	755/1027	73.5
< 75 years	581/895	64.9	1314/1831	71.8
≥ 75 years	80/115	69.6	187/242	77.3
Race				
White	468/681	68.7	1113/1454	76.5
Asian	164/290	56.6	340/554	61.4
Region				
North America	386/513	75.2	995/1222	81.4
Europe	68/135	50.4	121/226	53.5
Other Regions Combined	207/362	57.2	385/625	61.6

Table 121: Overall Summary of SAE - Pool 1

	Pool 1			
	Placebo N=1010		Total Istradefylline N=2073	
Subgroup	**n/N_1**	**%**	**n/N_1**	**%**
Gender				
Female	11/422	2.6	33/842	3.9
Male	20/588	3.4	54/1231	4.4
Age				
< 65 years	15/539	2.8	27/1046	2.6
≥ 65 years	16/471	3.4	60/1027	5.8
< 75 years	26/895	2.9	69/1831	3.8
≥ 75 years	5/115	4.3	18/242	7.4
Race				
White	25/681	3.7	59/1454	4.1
Asian	5/290	1.7	24/554	4.3
Region				
North America	15/513	2.9	52/1222	4.3
Europe	6/135	4.4	10/226	4.4
Other Regions Combined	10/362	2.8	25/625	4.0

Table 122: Summary of TEAEs, SAEs, TEAEs Leading to Discontinuation, and Severe TEAEs by Subgroup - Pool 2

		Total Istradefylline N=2132 n (%)			
Subgroups	**N_1**	**TEAEs**	**Serious TEAEs**	**TEAEs Leading to Discontinuation**	**Severe TEAEs**
Gender					
Female	816	739 (90.6)	162 (19.9)	110 (13.5)	203 (24.9)
Male	1316	1174 (89.2)	320 (24.3)	194 (14.7)	355 (27.0)
Age					
< 65 years	1133	1009 (89.1)	207 (18.3)	133 (11.7)	274 (24.2)
≥ 65 years	999	904 (90.5)	275 (27.5)	171 (17.1)	284 (28.4)
< 75 years	1690	1690 (89.3)	398 (21.0)	251 (13.3)	481 (25.4)
≥ 75 years	240	223 (92.9)	84 (35.0)	53 (22.1)	77 (32.1)
Race					
White	1639	1472 (89.8)	411 (25.1)	261 (15.9)	505 (30.8)
Asian	429	387 (90.2)	52 (12.1)	27 (6.3)	34 (7.9)
Region					
North America	1300	1255 (96.5)	387 (29.8)	234 (18.0)	491 (37.8)
Europe	252	166 (65.9)	26 (10.3)	25 (9.9)	23 (9.1)
Other Regions Combined	580	492 (84.8)	69 (11.9)	45 (7.8)	44 (7.6)

Age

In Pool 1 and Pool 2 the incidence of overall TEAEs by treatment group was similar across the age subgroups (<65, ≥65, <75, and ≥75 years).

However, serious TEAEs were more frequent in older subjects (≥ 65 years and ≥ 75 years) than in younger subjects (<65 years).

In addition, the incidence of severe TEAEs in the total istradefylline group, relative to the placebo group, was proportionately, greater for older subjects (≥ 65 years and ≥ 75 years) than for younger subjects (<65 years) in Pool 1. In Pool 2, severe TEAEs were also reported more often for older subjects than for younger subjects.

The incidence of TEAEs leading to discontinuation was higher with istradefylline treatment, compared with placebo, but was similar across the age subgroups in Pool 1. In Pool 2, TEAEs leading to discontinuation were reported more often for older subjects (≥ 65 years and ≥ 75 years) than for subjects < 65 years.

Overall, subjects ≥ 75 years had the highest incidence of Common TEAEs in both placebo and istradefylline groups (69.6% and 77.3%, respectively) compared with subjects < 65 years (67.2% and 71.3%, respectively) and subjects ≥ 65 years (63.5% and 73.5%, respectively).

With the exception of dyskinesia and fall, the higher incidence in the total istradefylline group, relative to the placebo group, was similar for all 3 age categories.

Relative to placebo, dyskinesia was proportionately more frequent for istradefylline-treated older subjects (≥ 65 years and ≥ 75 years) compared with istradefylline-treated younger subjects (< 65 years).

For subjects ≥ 75 years, the incidence of fall was proportionately greater (and 3% greater) for istradefylline-treated subjects than for placebo-treated subjects compared with the subgroup of younger subjects.

Table 123: Common TEAEs by Age Group (< 65, ≥ 65, and ≥ 75 years) in Pool 1

	< 65 years		≥ 65 years		≥ 75 years	
Preferred Term [a]	Placebo N=539 n (%)	Total Istradefylline N=1046 n (%)	Placebo N=471 n (%)	Total Istradefylline N=1027 n (%)	Placebo N=115 n (%)	Total Istradefylline N=242 n (%)
Subjects with any TEAE	362 (67.2)	746 (71.3)	299 (63.5)	755 (73.5)	80 (69.6)	187 (77.3)
Dyskinesia	57 (10.6)	186 (17.8)	40 (8.5)	183 (17.8)	7 (6.1)	31 (12.8)
Nausea	26 (4.8)	81 (7.7)	20 (4.2)	71 (6.9)	7 (6.1)	19 (7.9)
Dizziness	21 (3.9)	53 (5.1)	21 (4.5)	65 (6.3)	5 (4.3)	15 (6.2)
Constipation	16 (3.0)	48 (4.6)	17 (3.6)	63 (6.1)	5 (4.3)	18 (7.4)
Fall	24 (4.5)	41 (3.9)	26 (5.5)	55 (5.4)	7 (6.1)	22 (9.1)
Insomnia	26 (4.8)	56 (5.4)	16 (3.4)	38 (3.7)	4 (3.5)	4 (1.7)
Parkinson's disease[b]	23 (4.3)	43 (4.1)	14 (3.0)	35 (3.4)	3 (2.6)	7 (2.9)
Viral upper respiratory tract infection	25 (4.6)	39 (3.7)	9 (1.9)	35 (3.4)	3 (2.6)	2 (0.8)
Headache	19 (3.5)	43 (4.1)	11 (2.3)	27 (2.6)	2 (1.7)	5 (2.1)
Back pain	18 (3.3)	40 (3.8)	11 (2.3)	30 (2.9)	3 (2.6)	7 (2.9)

Hepatic Impairment

Two studies in patients with hepatic impairment were performed.

Mild hepatic impairment (Child Pugh A)

Study 6002-016 was an open-label study of a single dose of 40 mg/day of istradefylline in subjects with mild hepatic impairment (Child Pugh A). Higher incidence of TEAEs was observed in subjects with mild hepatic impairment compared to subjects with normal hepatic function.

Moderate hepatic impairment (Child Pugh B)

Study 6002-US-016 was Phase I, Multicenter, Open-Label, Parallel-Group Study to Assess the Effect of Moderate Hepatic Impairment (Child-Pugh B Classification) on the Pharmacokinetics and Safety of KW-6002 (Istradefylline) in Cigarette Smokers and Nonsmokers Following Administration of 40 mg Istradefylline Once Daily for 14 Days.

Table 124: Overall Summary of TEAEs (Safety Population)

Category of Subjects	Hepatically Impaired Smokers N = 7	Healthy Smokers N = 7	Hepatically Impaired Nonsmokers N = 7	Healthy Nonsmokers N = 7
Subjects with any TEAE	1 (14.3)	2 (28.6)	6 (85.7)	3 (42.9)
Subjects with any serious TEAE	0	0	0	0
Subjects who discontinued because of TEAE	0	0	0	0
Subjects with any related TEAE[a]	1 (14.3)	2 (28.6)	6 (85.7)	3 (42.9)
Subjects with any severe TEAE	0	0	0	0
Subjects with TEAE leading to death	0	0	0	0

Renal impairment

Study 6002-US-015 was a Phase 1 open label study to evaluate the influence of severe renal impairment on single dose PK and safety of 40 mg/day istradefylline (6002-US-015-r-en).

There were 18 subjects who enrolled and completed the study; 6 with severe renal impairment, 6 with normal renal function (matched by age and gender to the renally impaired group) and 6 young healthy adult subjects. There were no deaths, serious or severe TEAEs during the study and none of the subjects discontinued because of a TEAE. Seven subjects experienced at least 1 TEAE (Renally Impaired Group: 3 subjects; Matched Healthy group: 1 subject; Young Healthy group: 3 subjects).

Immunological events

Table 125: TEASs by SOC and PT for Immune system disorders pool 1

	Placebo (N=1010)		Istradefylline 10 mg/day (N=153)		Istradefylline 20 mg/day (N=869)		Istradefylline 40 mg/day (N=896)		Istradefylline 60 mg/day (N=155)		Total Istradefylline (N=2073)	
stem Organ Class/ Preferred Term [a]	n(%) of Subjects	Total AE(n)	n(%) of Subjects	Total AE(n)	n(%) of Subjects	Total AE(n)	n(%) of Subjects	Total AE(n)	n(%) of Subjects	Total AE(n)	n(%) of Subjects	Total AE(n)
Immune System Disorders	1 (0.1%)	1	0	0	1 (0.1%)	1	2 (0.2%)	2	2 (1.3%)	2	5 (0.2%)	5
Drug Hypersensitivity	1 (0.1%)	1	0	0	0	0	2 (0.2%)	2	0	0	2 (0.1%)	2
Allergy To Arthropod Sting	0	0	0	0	0	0	0	0	1 (0.6%)	1	1 (<0.1%)	1
Anaphylactic Reaction	0	0	0	0	1 (0.1%)	1	0	0	0	0	1 (<0.1%)	1

Safety related to DDI and other interactions

The Effect of Concomitant Treatments for PD was analysed.

These analyses defined 5 categories, based on pharmacologic class: DAs; COMT-inhibitors; MAO-B inhibitors; any of 2 of DAs; COMT-inhibitors; MAO-B inhibitors; and all 3 of DAs / COMT-inhibitors / MAO-B inhibitors. Subjects were categorized based on antiparkinson medications taken at baseline. Since the

protocols did not allow changes in these background medications during the studies, these same baseline anti-parkinson medications were taken concomitantly with test drug during the trials.

Table 126: Subjects with TEAS Stratified by Concomitant Use of Dopamine Agonists - Pool 1

	Yes		No	
Preferred Term [a]	**Placebo N=767 n (%)**	**Total Istradefylline N=1589 n (%)**	**Placebo N=243 n (%)**	**Total Istradefylline N=484 n (%)**
Subjects with any TEAE	495 (64.5)	1158 (72.9)	166 (68.3)	343 (70.9)
Dyskinesia	75 (9.8)	302 (19.0)	22 (9.1)	67 (13.8)
Nausea	29 (3.8)	116 (7.3)	17 (7.0)	36 (7.4)
Dizziness	28 (3.7)	87 (5.5)	14 (5.8)	31 (6.4)
Constipation	18 (2.3)	82 (5.2)	15 (6.2)	29 (6.0)
Fall	38 (5.0)	81 (5.1)	12 (4.9)	15 (3.1)
Insomnia	29 (3.8)	71 (4.5)	13 (5.3)	23 (4.8)
Viral upper respiratory tract infection	26 (3.4)	64 (4.0)	8 (3.3)	10 (2.1)
Back pain	23 (3.0)	54 (3.4)	6 (2.5)	16 (3.3)
Parkinson's disease[b]	26 (3.4)	51 (3.2)	11 (4.5)	27 (5.6)
Headache	21 (2.7)	46 (2.9)	9 (3.7)	24 (5.0)

Table 127: Subjects with TEAS Stratified by Concomitant Use of MAO-B Inhibitors - Pool 1

	Yes		No	
Preferred Term [a]	**Placebo N=278 n (%)**	**Total Istradefylline N=547 n (%)**	**Placebo N=732 n (%)**	**Total Istradefylline N=1526 n (%)**
Subjects with any TEAE	174 (62.6)	365 (66.7)	487 (66.5)	1136 (74.4)
Dyskinesia	26 (9.4)	94 (17.2)	71 (9.7)	275 (18.0)
Nausea	9 (3.2)	35 (6.4)	37 (5.1)	117 (7.7)
Dizziness	7 (2.5)	21 (3.8)	35 (4.8)	97 (6.4)
Constipation	10 (3.6)	28 (5.1)	23 (3.1)	83 (5.4)
Fall	6 (2.2)	21 (3.8)	44 (6.0)	75 (4.9)
Insomnia	8 (2.9)	20 (3.7)	34 (4.6)	74 (4.8)
Parkinson's disease[b]	11 (4.0)	13 (2.4)	26 (3.6)	65 (4.3)
Viral upper respiratory tract infection	14 (5.0)	15 (2.7)	20 (2.7)	59 (3.9)
Headache	8 (2.9)	16 (2.9)	22 (3.0)	54 (3.5)
Back pain	6 (2.2)	19 (3.5)	23 (3.1)	51 (3.3)

Table 128: Subjects with TEAS Stratified by Concomitant Use of COMT Inhibitors - Pool 1

	Yes		No	
Preferred Term [a]	**Placebo N=308 n (%)**	**Total Istradefylline N=809 n (%)**	**Placebo N=702 n (%)**	**Total Istradefylline N=1264 n (%)**
Subjects with any TEAE	209 (69.7)	630 (77.9)	452 (64.4)	871 (68.9)
Dyskinesia	31 (10.1)	183 (22.6)	66 (9.4)	186 (14.7)
Nausea	16 (5.2)	61 (7.5)	30 (4.3)	91 (7.2)
Dizziness	13 (4.2)	47 (5.8)	29 (4.1)	71 (5.6)
Constipation	12 (3.9)	48 (5.9)	21 (3.0)	63 (5.0)
Fall	20 (6.5)	41 (5.1)	30 (4.3)	55 (4.4)
Insomnia	16 (5.2)	42 (5.2)	26 (3.7)	52 (4.1)
Parkinson's disease[b]	12 (3.9)	38 (4.7)	25 (3.6)	40 (3.2)
Viral upper respiratory tract infection	9 (2.9)	32 (4.0)	25 (3.6)	42 (3.3)
Headache	14 (4.5)	27 (3.3)	16 (2.3)	43 (3.4)
Back pain	7 (2.3)	28 (3.5)	22 (3.1)	42 (3.3)

Dyskinesia was reported more frequently in istradefylline-treated subjects who received concomitant Treatments for PD with Dopamine agonists and COMT Inhibitors (19.0%, 22.6 % respectively) compared with istradefylline-treated subjects who did not receive concomitant these treatments (13.8%, 14.4% respectively). There was no difference in the frequency of dyskinesia between istradefylline-treated subjects who received MAO-B inhibitors versus those without such treatment. Dyskinesia was also more frequent in patients concomitantly receiving 2 or 3 medications for Parkinson's Disease.

Discontinuation due to AES

Treatment-emergent AEs leading to discontinuation occurred in 6.5% of subjects in the total istradefylline group and 5.2% of subjects in the placebo group with incidences of 5.6% and 7.3% in the istradefylline 20 mg/day and 40 mg/day groups.

Pool 1 (Double-blind, Placebo-controlled Fixed-dose Studies)

Table 129: TEAS Leading to Study Discontinuation Reported for ≥ 0.2% of Subjects in the Total Istradefylline Group by Preferred Term: Pool 1

		Istradefylline				
	Placebo N=1010 n (%)	**10 mg/day N=153 n (%)**	**20 mg/day N=869 n (%)**	**40 mg/day N=896 n (%)**	**60 mg/day N=155 n (%)**	**Total N=2073 n (%)**
Subjects with any TEAE leading to discontinuation from the study	53 (5.2)	5 (3.3)	49 (5.6)	65 (7.3)	16 (10.3)	135 (6.5)
Preferred Term[a]						
Dyskinesia	7 (0.7)	0	7 (0.8)	13 (1.5)	7 (4.5)	27 (1.3)
Parkinson's disease[b]	8 (0.8)	1 (0.7)	5 (0.6)	4 (0.4)	1 (0.6)	11 (0.5)
Nausea	1 (0.1)	1 (0.7)	1 (0.1)	4 (0.4)	4 (2.6)	10 (0.5)
Hallucination	1 (0.1)	0	2 (0.2)	6 (0.7)	0	8 (0.4)
Anxiety	0	0	2 (0.2)	5 (0.6)	0	7 (0.3)
Dizziness	2 (0.2)	0	1 (0.1)	5 (0.6)	0	6 (0.3)
Confusional state	1 (0.1)	1 (0.7)	1 (0.1)	3 (0.3)	0	5 (0.2)
Blood creatine phosphokinase increased	0	0	1 (0.1)	2 (0.2)	1 (0.6)	4 (0.2)
Insomnia	4 (0.4)	1 (0.7)	3 (0.3)	0	0	4 (0.2)
Muscle rigidity	1 (0.1)	0	2 (0.2)	2 (0.2)	0	4 (0.2)
Tremor	2 (0.2)	0	1 (0.1)	2 (0.2)	1 (0.6)	4 (0.2)

207.205. TEAEs leading to discontinuation, such as anxiety (20 mg 0.2% [2] and 40 mg 0.6% [5] vs. 0), blood creatine phosphokinase increased (20 mg 0.1% [1] and 40 mg 0.2% [2] vs. 0), ALT and AST increased (2 [0.2%] subjects in the 40 mg dose group v. 0), Blood alkaline phosphatase increased (3

[0.3%] subjects in the 40 mg dose group v. 0), occurred in subjects treated with the proposed doses of istradefylline but in none of those included in the placebo groups.

Pool 2 (Open-label, Long-term Studies)

Overall, 14.3% of subjects in the total istradefylline group discontinued from a study because of a TEAE.

Table 130: TEAS Leading to Study Discontinuation in ≥ 0.2% of Subjects in the Total Istradefylline Group by Preferred Term: Pool 2

	Total Istradefylline (20-60 mg/day) N=2132 n (%)
Subjects with any TEAE leading to discontinuation from the Study	304 (14.3)
Preferred Term[a]	
Parkinson's disease[b]	46 (2.2)
Dyskinesia	32 (1.5)
Hallucination	22 (1.0)
Psychotic disorder	10 (0.5)
Confusional state	9 (0.4)
Dizziness	7 (0.3)
Delusion	6 (0.3)
Atrial fibrillation	6 (0.3)
Pneumonia	6 (0.3)
Fall	5 (0.2)
Hallucination, visual	4 (0.2)
Nausea	4 (0.2)
Prostate cancer	4 (0.2)
Squamous cell carcinoma of skin	4 (0.2)
Constipation	3 (0.2)
Depression	3 (0.2)
Freezing phenomenon	3 (0.2)
Gastroenteritis	3 (0.2)
Myocardial infarction	3 (0.2)

Post marketing experience

Istradefylline was first approved with the brand name NOURIAST in Japan on 25 March 2013. Istradefylline was approved as NOURIANZ in the United States on 27 August 2019, at the same doses currently proposed (20 and 40 mg/day), for use as adjunctive treatment to L/C in adult patients with PD experiencing "OFF" episodes.

As of 31 May 2019, the cumulative post-marketing exposure estimate for istradefylline in Japan is approximately 63,500 patients. While NOURIANZ was approved in US in August 2019, there are no postmarketing data from the US at this time.

AE Reported in Post-marketing Setting

A total of 1580 AEs in 997 cases were retrieved; 364 AEs (23.0%) were serious and 1216 AEs (77.0 %) were non-serious. Adverse events were reported in non-interventional studies (1028 AEs), spontaneous reports (442 AEs), spontaneous literature (57 AEs), Investigator initiated study reports (34 AEs), non-interventional study literature (18 AEs), and study literature (1 AE).

The SOCs containing the largest numbers of reported AEs were nervous system disorders (336/1580 events, 21.3 %), psychiatric disorders (261/1580 events, 16.5%), injury, poisoning and procedural complications (216/1580 events, 13.7 %) and gastrointestinal disorders (158/1580 events, 10.0 %)

Table 131: Most Frequently Reported Post-Marketing Events by Seriousness and Relatedness

	Non-Serious			Serious			Total Serious and Non-serious	Overall AEs ≥ 0.5% Serious and Non-Serious (%)
Preferred Term	Not Related	Related	Total	Not Related	Related	Total		
Dyskinesia	13	134	147	-	3	3	150	9.5
Hallucination	18	71	89	2	5	7	96	6.1
Fall	44	2	46	6	3	9	55	3.5
Somnolence	4	45	49	-	-	-	49	3.1
Hallucination, visual	8	27	35	-	2	2	37	2.3
Contusion	27		27	3	-	3	30	1.9
Constipation	7	20	27	2	-	2	29	1.8
Dizziness	5	24	29	-	-	-	29	1.8
Off label use	24	4	28	-	-	-	28	1.8
Nausea	7	19	26	-	-	-	26	1.6
Nasopharyngitis	19	2	21	-	-	-	21	1.3
Insomnia	6	13	19	-	-	-	19	1.2
Pneumonia	2		2	15	-	15	17	1.1
Delusion	6	7	13	1	3	4	17	1.1
Dysphagia	10	2	12	3	1	4	16	1.0
Abdominal discomfort	3	9	12	-	-	-	12	0.8
Decreased appetite	3	8	11	-	1	1	12	0.8
Tremor	2	8	10	1	1	2	12	0.8
Delirium	1	1	2	3	7	10	12	0.8
Rash		12	12	-	-	-	12	0.8
Blood creatine phosphokinase increased	4	7	11	-	-	-	11	0.7
Back pain	7	2	9	2		2	11	0.7
Pneumonia aspiration	-	-	-	10	1	11	11	0.7
Hepatic function abnormal	2	5	7	-	3	3	10	0.6
Parkinson's disease	1	1	2	7	1	8	10	0.6
Abdominal pain upper	3	5	8	-	1	1	9	0.6
Oedema peripheral	4	4	8	-	1	1	9	0.6
Diarrhea	3	4	7	-	1	1	8	0.5
Vomiting	3	3	6	1	1	2	8	0.5

(Table continues)

Preferred Term	Non-Serious			Serious			Total Serious and Non-serious	Overall AEs ≥ 0.5% Serious and Non-Serious (%)
	Not Related	Related	Total	Not Related	Related	Total		
Skin laceration	7	-	7	1	-	1	8	0.5
Spinal compression fracture	1	1	2	6	-	6	8	0.5
Pollakiuria	3	5	8	-	-	-	8	0.5

Table 132: Most Frequently Reported Post-Marketing SAE by Relatedness

System Organ Class Preferred Term	Serious Adverse Events		
Cardiac disorders	**Not Related**	**Related**	**Total**
Cardiac failure	2	1	3
Cardiac failure acute	1	2	3
Myocardial infarction	2	1	3
Gastrointestinal disorders			
Dysphagia	3	1	4
Ileus paralytic	2	1	3
General disorders and administration site conditions			
Death	3	1	4
Gait disturbance	3	1	4
Sudden death	2	3	5
Hepatobiliary disorders			
Cholecystitis	2	1	3
Hepatic function abnormal	-	3	3
Infections and infestations			
Pneumonia	15	-	15
Injury, poisoning and procedural complications			
Contusion	3	-	3
Fall	6	3	9
Femoral neck fracture	3	1	4
Femur fracture	3	1	4
Lumbar vertebral fracture	3	-	3
Rib fracture	3	-	3
Spinal compression fracture	6	-	6
Subdural hematoma	3	-	3
Musculoskeletal and connective tissue disorders			
Rhabdomyolysis	2	4	6
Neoplasms benign, malignant and unspecified (incl cysts and polyps)			
Gastric cancer	4	1	5
Nervous system disorders			
Dementia	3	-	3
Dyskinesia	-	3	3
Parkinson's disease	7	1	8
Sudden onset of sleep	1	2	3
Psychiatric disorders			
Delirium	3	7	10
Delusion	1	3	4
Depression	2	2	4
Hallucination	2	5	7
Suicide attempt	1	2	3
Respiratory, thoracic and mediastinal disorders			
Pneumonia aspiration	10	1	11

Amongst the most frequently reported AEs in post-marketing setting, dyskinesia, hallucinations, constipation, dizziness, nausea and vomiting are recognized as adverse reactions with istradefylline based on clinical trial data.

A review of the remaining most frequently reported AEs revealed that the majority of events represented PD symptomatology; PTs of PD (the majority of these events reported verbatim of aggravation of PD), tremor, dysphagia, pollakiuria, or known comorbidities in patients with PD (PTs of fall and resulting injuries, pneumonia; pneumonia aspiration; sleep disorders [somnolence, insomnia], and psychiatric disorders [delirium, delusion]).

Off-label use was also 1 of the most frequently reported PTs (n=28, 1.8% of all AEs). The types of off-label use included use in unapproved indication, unapproved posology, and method of administration. While AEs were reported in some cases, no information indicative of a new risk with off-label use was observed.

During routine signal detection activities, a safety signal of rash with istradefylline was detected from post-marketing data in Japan. The assessment of the signal of rash concluded that, there is sufficient evidence presented in the cases of rash to consider it an ADR with istradefylline. Key factors supporting this conclusion include the available data on positive dechallenge, and plausible latency.

2.6.1. Discussion on clinical safety

Safety data pooling

The safety data from twenty-four phase 2/3 studies were organised into 5 pools. Pools 1 and 2 are considered to be the most relevant in the context of this application whereas Pool 3 (safety data from Phase 2 Pilot Studies), Pool 4 (studies of other indications) and Pool 4a (studies of istradefylline as monotherapy in PD) are considered as supportive only.

Pool 1 includes 8 DB, randomised, placebo-controlled fixed-dose studies of 12- or 16-weeks duration. All subjects enrolled to these studies were receiving levodopa, in combination with a peripheral dopa-decarboxylase inhibitor (carbidopa or benserazide), and most were also receiving other additional standard anti-parkinson medication(s). In Pool 1 2073 patients were exposed to istradefylline. Most of the subjects (1765 of 2073 subjects) in Pool 1 who received istradefylline were treated with the proposed dosing regimen of 20 or 40 mg/day.

Pool 2 includes 5 OL, single arm, long-term extension studies of istradefylline in subjects with idiopathic PD and motor response complications while taking levodopa therapy. Pool 2 provides the safety data from 2132 subjects, including 1345 subjects treated with istradefylline for at least 1 year. It needs to be highlighted however, that the design of these extension studies (single arm, no comparator) limits the ability for the causality assessment.

Other Pools (Pool 3 and Pool 4) included smaller studies. Pool 3 includes two early PD studies in which 149 patients were exposed istradefylline. Pool 4 includes 6 studies of istradefylline as monotherapy in PD (2 studies), major depressive disorder (2 studies), and RLS (restless leg syndrome) (2 studies). In all these studies 490 patients were treated with istradefylline.

Exposure

The total exposure to istradefylline for subjects in a Phase 2 or 3 study was 3079 PYs; 86% of these subjects (2997/3501 subjects; 3021 PYs) received istradefylline as adjunctive therapy for PD. A total of 1215 subjects were exposed to istradefylline for longer than 12 months and 413 subjects were exposed to istradefylline for longer than 24 months as adjunctive therapy for PD.

This exposure fulfils the requirements of the ICH E1 guideline. A total of 252 subjects (50.6%) in the istradefylline 20 mg/day and 1009 subjects (68.9%) in the istradefylline 40 mg/day received continuous treatment with istradefylline for at least one year.

The number of subjects aged ≥75 years old is relatively small (343/2997, 11.4%) and particularly patients ≥85 years (11, 0.4%) are under-represented.

Common TEAEs

Treatment-emergent Adverse Events were reported more frequently in patients receiving istradefylline treatment. In Pool 1, 72.4% of subjects in the total istradefylline group experienced at least 1 TEAE compared to 65.4% of subjects in the placebo group. The incidences of subjects with any TEAE were similar in the istradefylline 20 mg/day and 40 mg/day groups (70.7% and 70.1% respectively), and higher in the istradefylline 10 mg/day and 60 mg/day group (82.4% and 85.8% respectively).

In Pool 1, the SOC with the most frequently reported TEAEs was nervous system disorders, for which events were reported for 34.1% of subjects in the total istradefylline group and 26.8% of subjects in the placebo group. Other SOCs with TEAEs reported with at least a 10% incidence were gastrointestinal disorders, psychiatric disorders, investigations, musculoskeletal and connective tissue disorders, and infections and infestations. For gastrointestinal disorders, psychiatric disorders the frequency of TEAEs was higher the istradefylline group than in the placebo group.

The most frequently reported TEAEs for subjects in the istradefylline group were dyskinesia, nausea, dizziness, constipation, fall, and insomnia.

In Pool 2 89.7% of subjects reported any TEAEs. Also in Pool 2, the SOC with the most frequently reported TEAEs was nervous system disorders (as 64% reported nervous system disorders). This was followed by the SOCs: psychiatric disorders, gastrointestinal disorders, musculoskeletal and connective tissue disorders, infections and infestations. The most frequently reported TEAEs in total in Pool 2 were dyskinesia (36.3%), (worsening symptoms of) PD (24.3%), fall (16.2%), insomnia (15.0%), and constipation (15.0%)

For dyskinesia the difference between the istradefylline group and placebo was most apparent (8% difference). In Pool 1 the incidence of dyskinesia was greater in the total istradefylline group (17.8%) compared with the placebo group (9.6%), with similar incidences of dyskinesia in the istradefylline 20 mg/day (16.1%) and istradefylline 40 mg/day (17.7%) groups. Dyskinesia was also more frequently reported in other Pools.

For other TEAEs in Pool 1 the difference between groups was smaller i.e for nausea there was 2% differences, for dizziness 1 % difference, constipation 1.6 % difference and hallucination 1% difference.

The applicant proposed to list dyskinesia, nausea, dizziness, constipation, hallucination and vomiting as AEs. A better description of the different types of hallucinations was requested as well as a discussion for monitoring the possible onset of these ADRs and for considering dose reduction or treatment interruption in case a patient presents hallucinations**.**

It is noted that in the FDA label for istradefylline, diarrhea, blood urea increased and Blood alkaline phosphatase increased are listed. Therefore, the applicant was asked to discuss and consider including these ADRs**.** Based on the data provided by the applicant it could be agreed that in respect to diarrhea, blood urea increased and blood alkaline phosphatase increased the difference between the treatment groups was small.

Deaths

The provided data do not indicate that there is a higher risk of death of patients receiving treatment with istradefylline however some clarifications are required.

Nine subjects died in Pool 1. There we 5 deaths in the placebo group and 4 deaths in the treatment group. Only pneumonia was reported twice. Other TEAEs with an outcome of death were reported only once.

Thirty-two subjects died in Pool 2.

In Pool 2 the most frequently reported TEAEs with an outcome of death were pneumonia (7 subjects [0.3%]), aspiration pneumonia (4 subjects [0.2%]), (worsening symptoms of) PD (3 subjects [0.1%]), and cardiac arrest (3 subjects [0.1%]). In addition, 2 patients had cardio – respiratory arrest, and 2 patients had sudden death (Subject077-0015 and Subject 077-0010). All patients with cardiovascular deaths had a significant history of cardiovascular disease.

The applicant was requested to comment whether the frequency of deaths (especially sudden death) in Pool 2 is in line with the frequency observed in this population of patients. In addition, it is noted that the percentage of patients who died differs between studies. The higher frequency of deaths was reported in study 6002-US-025 (3.2%) and in study 6002-018 (2%). On the other hand, in studies 6002-US-007 and 6002-INT-001 only 0.6% of patients died. In study 6002-010 no deaths were reported. The applicant provided the requested clarifications on some of the deaths of patients treated with istradefylline potentially related to cardiovascular or respiratory disorders.

Serious TEAEs

In general, the number of patients who experienced serious TEAEs in Pool 1 was small however, the percentage of such patients was slightly higher in the istradefylline group (4.2%) as compared to the placebo group (3.1%). More serious TEAEs were reported in patients receiving 40 mg dose as compared to those treated with 20 mg.

The most frequently reported serious TEAEs were falls and pneumonia and for those there was no significant differences between istradefylline and placebo.

For the rest of serious TEAEs it is difficult to make any firm conclusion as only single cases of serious TEAEs were reported.

Safety profile depending on the dose (20 mg or 40 mg)

There are concerns that the higher proposed dose of istradefylline (40 mg/day) could be slightly less safe as compared to the lower dose especially in relation to the development of serious and severe TEAEs after a long term treatment with istradefylline.

The percentage of subjects with any TEAE was similar in the istradefylline 20 mg/day and 40 mg/day groups (70.7% and 70.1% respectively).

Serious TEAEs in Pool 1 were reported in 3.9% in the 20 mg group and 4.8% in the 40 mg group. The overall exposure-adjusted incidence rates (EAIR) for serious TEAEs (n$\geqslant$2) in Pool 1 were lower in the istradefylline 40 mg/day as compared to the 20 mg/day group (0.28 vs. 0.33, respectively). Severe TEAEs in Pool 1 were reported in 7.6 % of subjects in the 20 mg group and 7.9 % subjects in the 40 mg group. The overall EAIRs for severe TEAEs (n=$\geqslant$2) in Pool 1 were lower in the 40 mg/day group than in the 20 mg/day group (0.58. vs. 0.64, respectively) with a small difference.

In Pool 2 however serious and severe occurred more frequently in patients receiving the higher istradefylline dose.

In Pool 2 serious TEAEs were reported in 16.1% of subjects receiving 20 mg dose as compared to 25.9% subjects receiving the 40 mg dose. The incidence per patient-year exposure of serious TEAEs was 0.27 in the 20 mg/day group and 0.34 in the 40 mg/day group in Pool 2.

Severe TEAEs were also reported more frequently (in 28.3%) in the 40 mg treatment group as compared to 19.5% in the 20 mg treatment group and the difference between the treatment groups was particularly seen in relation to severe TEAEs considered to be adverse drug reactions, e.g., hallucinations, nausea and vomiting. In long term studies, the overall EAIR of severe TEAEs was numerically higher in the 40 mg/day group (0.46) as compared to the 20 mg/day group (0.39).

Based on the presented results it could be concluded that in respect to serious, severe TEAEs and ADRs the differences in EAIRs between the 20 mg/day group and the 40 mg/day group were small. Nevertheless, it seems that during a long-term treatment the 40 mg dose could be associated with a slightly less favorable safety profile as serious and severe TEAEs were reported more frequently in patients receiving higher dose.
As indicated in the efficacy sections of this assessment report, that there is no consistent evidence of greater efficacy with the 40mg compared to 20mg dose. For this reason, the justification for the use of the 40mg dose is being questioned.

In Pool 1 data the EAIR for grouped hallucination was numerically higher in istradefylline 40 mg/day vs 20 mg/day dose (0.22 vs 0.15 respectively).The opposite observation was present in Pool 2 data where the EAIR for hallucination was numerically higher in 20 mg/day vs 40 mg/day group (0.17 vs 0.15).

It can be agreed with the applicant that the differences between the treatment groups was small.

Long-term AE

The EAIRs of TEAEs reported for ≥ 10% of subjects in the total istradefylline group in Pool 2 was compared with the corresponding rates for subjects in the placebo group in Pool 1. With the exception of TEAEs of (worsening symptoms of) PD, the EAIRs of these TEAEs in the total istradefylline group in Pool 2 were similar to or lower than those reported for subjects in the placebo group in Pool 1. The applicant was requested to compare the exposure-adjusted incidence rates for TEAEs reported in less than 10% subjects, especially for Serious TEAEs. The relevant discussion was provided. The applicant considered that the comparison of EAIRs for TEAEs >1.0% for Pool 2 with placebo EAIRs in Pool 1 did not identify any new safety concerns associated with long-term use of istradefylline. Taking into consideration a small number of events especially in the placebo group, the limitation of this comparison is acknowledged. For some imbalances the observed differences between Pool 2 and placebo in Poo1 it could be a chance finding.

However, further discussion was required for differences in relation TEAE of blood glucose increased, psychiatric disorders and cognitive impairment.

Some small imbalances between the Exposure-adjusted incidence rates (EAIRs) for in Pool 2 as compared to the exposure-adjusted incidence rates in placebo in Pool 1 were noted for memory impairment, cognitive disorder and confusion state. Therefore, further discussion and justifications were requested. The applicant clarified that the EAIR for memory impairment is 0.01 each in the placebo and istradefylline 40 mg/day groups and 0.02 in the istradefylline 20 mg/day group. The applicant considered that the difference in EAIR of 0.01 suggests incidental rather than a causal occurrence. Confusional state has already been included as an ADR .

Use in patients with cognitive impairment is proposed as missing information. The applicant was required to discuss and justify the inclusion of this safety concern. It was clarified that no definitive conclusion about the safety profile of istradefylline in patients with cognitive impairment could be made.

The TEAE of blood glucose increased was considered as ADR.

Adverse Events of Special Interest (AEoSIs)

The applicant identified a number of AEoSIs which were discussed and reviewed separately.

As regard to orthostatic hypotension, in Pool 1, orthostatic hypotension-related AEs were reported in the 9.4% and 7.4% of patients treated with istradefylline 20 mg/day and 40 mg/day, respectively, versus 6.2% of placebo patients. The most frequently TEAE related to orthostatic hypotension was dizziness (20 mg 5.1% and 40 mg 5.9% vs. 4.2% in the placebo group), but also the specific TEAE orthostatic hypotension was reported with a higher incidence in the istradefylline group, doubled compared to placebo (2.2% and 0.7% vs. 1.1%). The applicant was asked to consider to include orthostatic hypotension as an ADRs. However, based on the submitted justification, the overall incidences of subjects who met the orthostatic hypotension criteria were overall similar between placebo and istradefylline dose groups of 20 mg and 40 mg in Pool 1, the frequency of orthostatic hypotension doubling the frequency of the placebo patients (2.2% vs. 1.1%) was observed in the 20 mg dose group but not at 40 mg of istradefylline (0.7%) which can stand against a causal relationship with istradefylline. Furthermore, it is acknowledged that patients with PD can present orthostatic hypotension as a consequence of the cardiac autonomic dysfunction related to the underlying disease. In addition, also concomitant anti-PD can cause orthostatic hypotension. These aspects could represent confounding factors.

It is noted that in the 75 to 84 age-category, subjects in the istradefylline 20 mg/day (21.7%) and 40 mg/day (17.8%) groups were reported to have higher frequencies of the grouped term "postural hypotension, falls, black outs, syncope, dizziness, ataxia, and fractures" compared to the placebo group

(12.1%). The applicant considers however that the data do not suggest that istradefylline has less favourable safety profile in older subjects. Apart from dizziness, none of the AE listed in this grouped term are recognized as ADR with istradefylline. In addition, it was highlighted that the size of the 75 to 84-year-old group is approximately 3 times smaller than that of 65 to 74-year-old group and approximately 4 times smaller than the < 65-year-old group, which may have resulted in more variance in the observed incidence of TEAEs in 75 to 84-year old group. Limited data on use of istradefylline exits in subjects' ≥85 years old.

The incidence of TEAEs in the cardiac disorders SOC was higher in the placebo group (3.5%) and istradefylline 60 mg/day group (5.2%) than in the istradefylline 10 mg/day (2.0%), istradefylline 20 mg/day (3.0%) or istradefylline 40 mg/day (2.8%) groups. The frequency of any particular TEAE by PT was not higher in the istradefylline groups with exception of heart failure which was reported in 4 subjects receiving istradefylline in Pool 1 and 0 subjects receiving placebo. Left ventricular hypertrophy, and Tricuspid valve incompetence (20 mg and 40 mg 2 [0.2%] subjects each vs. none subject in the placebo group) were reported only in subjects receiving istradefylline compared with none subjects in the placebo group. Two events of myocardial infarction occurred in the istradefylline group (40 mg 2 [0.2%] vs. 1 [0.1%]) and were considered serious and both related to treatment (40 mg 2 [0.2%] vs. 0 in placebo). In the long-term open-label Pool 2, the most frequently reported cardiac TEAEs were increases in blood CK levels (3.7%) mostly of muscular origin except in 1 subject where it was of myocardial origin, and myocardial infarction (0.6%); the majority of myocardial infarction events were serious. In the monotherapy Pool 4a, 1 AE of angina unstable was reported in 1 subject treated with 40 mg/day of istradefylline. In the post-marketing setting (as of 31 May 2019), a total of 32 (2% of all AEs) cardiac AEs were reported, of them 23 were SAEs.

The corresponding EAIRs for these same AEs from Pool 2 were 0.01 or < 0.01 per PY of exposure with long-term istradefylline treatment.

However, considering the seriousness and relationship to istradefylline treatment of the cardiac AEs, their clinical relevance, and the mechanism of action of istradefylline that may have potential implications for cardiovascular system and acknowledging the existence of a number of confounding factors (age, comorbidities, concurrent medications) that limit the causality assessment, it is at present not possible to exclude a causal relationship between treatment with istradefylline and cardiovascular AEs.

The applicant have thoroughly discussed the cardiovascular AE-related issue based on both non-clinical and clinical available data. Given the above, the applicant concluded that these data do not support a clear direct relationship of istradefylline treatment with cardiac events. This is acknowledged; however, the applicant was requested to commit monitoring of any signal on cardiac events suggestive of potential causal relationship with istradefylline treatment in the post-marketing setting with consequent reporting and thorough evaluation in the PSUR**.**

Regarding respiratory TEAEs non clinical studies showed microscopic changes in the lung of rats, mice, and dogs treated with istradefylline compatible with inflammatory alterations. In Pool 1, the respiratory TEAEs occurring with higher frequency in the istradefylline group than the placebo group were cough (20 mg 1.0% and 40 mg 1.5% vs. 0.7%), dyspnoea (1.4% and 0.4% vs. 0.9%), upper respiratory tract inflammation (0.6% and 0.9% vs. 0.1%), and bronchitis (0.8% and 1.0% vs. 0.3%). The slightly higher frequency of the upper respiratory tract infections observed in Pool 1 is apparently mainly driven by upper respiratory tract inflammation term used for indicating acute upper respiratory tract inflammation. Respiratory TEAEs were the most frequently reported also in the long-term open-label Pool 2, and both upper and viral upper respiratory infections occurred with a higher incidence also with istradefylline monotherapy compared to placebo. Given the above and the biological plausibility related to the

istradefylline mechanism of action, the applicant was requested to consider upper respiratory tract inflammation as an ADRs.

Psychiatric disorders

As mentioned above, in Pool 1 TEAEs within the SOC Psychiatric disorders were reported more frequently in the istradefylline group (16.2%) as compared to those receiving placebo (11.4%). The highest frequency was seen in the highest dose group (24.5% in patients receiving 60 mg dose). TEAEs Anxiety had a slightly higher frequency in Pool 1 and in Pool 3.

Also serious TEAEs within the SOC Psychiatric disorders were reported with the higher frequency in the istradefylline group (10 cases in total) as compared to 1 case in the placebo group. The following serious TEAEs were reported: confusion sate, delirium, psychotic disorders, depression, disorientation, hallucination, persecutory delusion, psychiatric symptoms and suicidal attempt. It is noted that patients with psychotic illness or depression were excluded from all studies. Serious TEAEs of anxiety, confusional state, hallucination, persecutory delusion was also reported in patients receiving istradefylline in Pool 3 and also in this pool no serious TEAEs within the SOC Psychiatric disorders were reported in the placebo group.

From the requested more in-depth discussion on the AE anxiety, it was apparent that there were no clinically significant differences in the frequencies of these AEs between the 10 mg, 20 mg and 40 mg dose groups of istradefylline and placebo group; these data were supported by the exposure-adjusted incidence rates from the long-term Pool 2 in comparison with the placebo group of Pool 1.

As pointed out above, TEAEs of hallucinations has been already proposed by the applicant to be listed as ADR

Regarding psychotic disorders other than hallucinations, since in Pool 1, the TEAEs mania (3 [0.3%] subjects receiving 40 mg/day of istradefylline), agitation (20 mg 2 [0.2%] and 40 mg 3 [0.3%]), delirium (20 mg and 40 mg 2 each [0.2%]), and abnormal behaviour (20 mg 3 [0.3%]) were reported only in the proposed doses of istradefylline versus no cases in the placebo group, the applicant was asked to consider these psychotic disorders as ADRs. After an in-depth discussion, the applicant considers that, despite the slight imbalance between istradefylline and placebo in Pool 1, the incidence of psychotic disorders observed in the istradefylline groups was low and within the incidence expected in the PD population. However, taking into account all the available information and the potential biological plausibility of the mechanism of action, the applicant proposed psychotic disorders as an important potential risk in the RMP.

Psychotic disorders were considered as ADR with the frequency "uncommon" based on Pool 1 data, since a causal relationship with istradefylline treatment has been established. For this reason, psychotic disorders have been re-classified from an important potential to an important identified risk in the RMP. Since hallucinations are symptoms of psychotic disorders, they were merged under psychotic disorders rather than listed as a separate risk in the RMP, in accordance with GVP V guidelines; this is acceptable.

Impulse control disorder

In controlled studies (Pool 1), adverse reactions consistent with impulse control disorder (including hypersexuality, hyperphagia, impulse control, increased libido, gambling, increased appetite, and obsessive-compulsive thoughts/disorder) were observed in 0% of patients in the placebo group, and in 0.7%, 0.8%, 0.6%, and 0% of patients in the istradefylline 10, 20, 40, and 60 mg/day groups, respectively. Although the differences in incidence between istradefylline and placebo were small, all cases of impulse control disorder (n=13) occurred in istradefylline treated subjects.

Suicidality

There is some imbalance in the number of subjects with the AEoSI term suicidality between the treatment groups and placebo group (although a small number of events needs to be noted). In Pool 1 all 4 cases were seen in the istradefylline groups and there were no cases in the placebo group.

Pool 2 there were 13 cases within the Grouped Term of suicidality, 5 TEAEs were considered to be serious, and 2 subjects were reported as having treatment-related serious TEAEs. In postmarketing 3 suicidal attempts (3/63,500) have been reported.

Sleep disorders

The review of the Pool 1 data indicated some imbalances in the percentage patients who experienced sleep disturbances. Insomnia, abnormal dreams, sleep disorder occurred more frequently in patients treated with istradefylline as compared to those receiving placebo although a small number of the events is noted.

Taking into consideration imbalances seen in Pool 1 and direct pharmacological effect of istradefylline (A2a inhibition would be expected to increase alertness) the casual relationship between istradefylline treatment and sleep disturbances cannot be fully excluded.

The applicant agrees to consider insomnia and abnormal dreams as an ADR based on the approximately 1% higher frequency of these two TEAEs in either the istradefylline 20 mg/day or 40 mg/day groups vs placebo in Pool 1.

Abuse potential

The applicant performed Study 6002-017 to investigate abuse potential of istradefylline. In this study there was a trend towards signs of abuse potential of istradefylline.

Istradefylline doses of 40 mg, 80 mg, and 160 mg showed statistically greater effects than placebo on the primary endpoint of Drug Liking VAS E_{max} and some secondary endpoints of balance of effects, positive effects, stimulant effects and any effects.

The applicant argues that the mean differences for istradefylline are not consistent with clinically important abuse potential because they are less than the minimum ~15-20 point differences from placebo that are observed with drugs of abuse across multiple drug classes (Schoedel et al, 2012b). Further, the upper limit of the 95% CIs of differences in Drug Liking VAS E_{max} between all 3 istradefylline doses and placebo were <11 points; a proposed equivalence margin for determining clinically meaningful differences from placebo, based on a meta-analysis of human abuse potential studies (Chen & Bonson, 2013). However, no equivalence margin was pre-specified and justified in the study protocol.

Significant limitations in the completed non-clinical package for assessing drug dependency have been identified, specifically in relation to the completed study assessing the reinforcing effects of Istradefylline in Rhesus monkeys. Given such limitations, further weight was placed on the results of the completed clinical study. The applicant was requested to discuss and provided a comprehensive discussion and adequately justified that istradefylline has a low risk of abuse potential.

Brain vascular mineralisation

Histopathological examination of the rodent brains, revealed foci of mineralization which were usually evident in the walls of arterioles and small arteries primarily located in the basal ganglia and, more rarely, in other parts of the brain.

In rats, the foci of mineralisation were observed rarely in control animals but increased with dose and exposure duration in istradefylline treated rats.

The applicant's position is that the finding in rats is a species-specific effect, posing a low potential risk in terms of human safety (in mice the foci were observed in both control and istradefylline treated animals whereas in dogs no foci of vascular mineralization was observed).

Primary familial brain calcification (PFBC) is a rare neurological condition which provides a potential model of what might be identified as "primary" mineralization in the basal ganglia and other parts of the brain. The symptoms of this condition included dementia, psychosis, mood swings, loss of motor skills, rigidity, and spastic paralysis. Dystonia, athetosis and chorea have also been observed in some patients. In addition, Parkinsonian features such as tremors and rigidity, masklike facial expression, shuffling walk, and pill-rolling have been reported.

The applicant claims that the available safety data provide reassurance that this concern is not relevant to humans.

This is not fully agreed. The safety data from Pool 1 are considered to be of a limited value in the context of this safety concern as 3 months exposure to istradefylline in Pool1 is likely to be too short for the development of these brain changes.

The review of the Pool 2 or postmarking data is considered to be more relevant however, there are some limitations. Possible symptoms of brain vascular mineralization are likely to be difficult to be distinguished from symptoms of PD or AEs linked to PD medications. The applicant examined the post-marketing safety database for findings that might suggest development of foci of vascular mineralization in the brains of human patients is provided. However, this review was performed 5 years after authorization in Japan.

Therefore, although the preclinical signal is not very strong, the applicant was requested to further discuss this issue. In addition, the applicant agrees to add brain vascular mineralisation to the RMP as an important potential risk. Adverse events representing CNS symptomatology, which could potentially be associated with brain vascular mineralisation, will be performed as part of ongoing routine signal detection activities. "Brain vascular mineralization" was added as an important potential risk to the RMP. The applicant was requested to discuss and propose a more specific safety concern to be included in the RMP that reflects the possible clinical relevance of the animal studies findings. The applicant has re-named the important potential risk of 'Brain vascular mineralisation' to 'Movement, neurological or psychiatric disorders due to brain vascular mineralisation'. Movement disorders as those described in the applicant's response, are part of neurological disorders. As requested, this important potential risk was further re-named as follows: 'Neurological (mainly movement) disorders or psychiatric due to brain vascular mineralisation'.

Haematology

There were no clinically meaningful mean changes from Baseline to Endpoint in the haematology analytes in Pool 1 in the treatment groups as compared to the placebo group. Shifts from normal-to-low or normal-to-high were observed in < 10% of subjects in any dose group for all of the haematology analytes in Pool 1. For Pool 2 some clarification was required in relation to changes in haemoglobin levels as well as in relation to the trend towards a slightly higher incidence of decrease in haematocrit and increase in leucocyte numbers in the istradefylline group compared with placebo. It is acknowledged that in Pool 1 there were discrepancies between PCS increased leukocytes ≥1.2 x ULN -which was reported in 2% in the istradefylline 40 mg/day group compared with 1% each in the istradefylline 20 mg/day group and placebo- and the TEAE of White blood cell count increased -which was reported more frequently in the 20 mg/day dose group than in the 40 mg/day dose group. Similarly, no. difference was found among the treatment groups in terms of PCS neutrophils increased, while TEAE for neutrophil increased were reported only in istradefylline 20 and 40 mg/day groups. Furthermore, no relevant differences were observed among the treatment groups for the TEAE leucocytosis.

It is acknowledged that the presence of these discrepancies does not allow to draw definite conclusions on the relationship between increase of white blood count and istradefylline treatment. Moreover, it is agreed that, as pointed out in the question, increased white blood count in 40 mg/day dose group could be related to the higher incidence of infections, particularly in terms of upper respiratory tract inflammation and this AE, as requested, has been included by the applicant as an ADR. Finally, it is acknowledged that the observed differences were not clinically relevant.

Chemistry

Liver enzyme elevations

In pivotal studies AE of increased liver enzymes was reported 2.0% in the placebo group and 1.3%, 1.2%, 2.5%, and 2.6% in the istradefylline 10, 20, 40, and 60 mg/day groups, respectively showing slightly higher frequency in patients receiving higher doses. Cases of serious AEs of increased liver enzymes were reported exclusively in the istradefylline treatment groups and also only the istradefylline treatment, increased liver enzymes led to discontinuation. No clear case of Hy's law was reported. No cases of hepatic failure were reported.

More cases of increased liver enzymes in the istradefylline treatment groups were seen in the review of the laboratory data.

In the E-R analysis of istradefylline (2018), which is detailed in the PK section this report, there was an increase in liver enzyme elevations with increasing istradefylline exposure. The incidence of liver enzyme elevations increased from 0.8% in the lowest quartile of exposure to 4.7% in the highest quartile of exposure. However, there were no serious liver enzyme elevations.

Based on the totality of data it seems that istradefylline treatment could have some potential to increase level of liver enzymes. The applicant was requested to further discuss this safety concern especially and consider monitoring of liver enzymes in the postmarking setting. The applicant concluded that the available data do not support the conclusion that istradefylline causes liver injury or elevation in liver function tests. However, the applicant acknowledges some uncertainties with regards to non-clinical findings and the population pharmacokinetic analysis.

Pancreatic enzyme

Pancreatic cell vacuolation and apoptosis/single cell necrosis were reported in non-clinical studies. However, analysis of clinical laboratory results does not indicate that istradefylline is associated with elevations of serum amylase or lipase activity or of trypsin-like immunoreactivity. In the post-marketing data there was only two cases of events of amylase increase which were reported in patients receiving treatment for more than 6 months. There was no case of pancreatitis. However, the evaluation of pancreatic atrophy, such as that observed in non-clinical studies, cannot rely only on serum biomarkers, and needs imaging examinations of the pancreas, that were not performed in the istradefylline clinical studies.

The applicant pointed out that, although imaging studies were not performed during the clinical development program of istradefylline, an evaluation of the AEs related to chronic pancreatitis and pancreatic atrophy in the form of diabetes was performed. In Pool 1, diabetes mellitus was reported in 1 case each in the 10 mg and 20 mg dose groups and in 3 cases in the 40 mg group vs. 1 case in the placebo group. One 20 mg subject and two 40 mg subjects had previous history if diabetes which however worsened during the treatment with istradefylline, the other patient receiving 40 mg of istradefylline and the patients treated with 10 mg/day developed diabetes mellitus in a short-term (after 77 and 57 days, respectively). Pool 2 data showed differences when compared with the placebo group of Pool 1, for the higher number of subjects who reported to have developed diabetes mellitus or worsening of pre-existing diabetes observed in the 40 mg istradefylline group (11 [0.8%] vs. 1 [0.1%]

in the placebo group). None of these events was serious, only one was considered possibly related to treatment, no action was taken regarding istradefylline treatment, and the event resolved with sequelae. In the post-marketing setting, 4 cases of diabetes mellitus were reported. Of them, 3 were considered serious worsening of pre-existing diabetes or development of diabetes in pre-diabetes occurring 22, 12, or 2 months after starting treatment with istradefylline 20 mg/day. In one of these cases istradefylline was discontinued and the event was recovering. All events resolved but one. One of the 4 cases was non-serious worsening of diabetes after approximately 8.5 months of therapy with istradefylline 20 mg/day.

It is acknowledged that it is not possible to conclude that these cases of diabetes mellitus are indicative of patients with chronic pancreatitis or atrophy. However, all the data reported above on the AE diabetes mellitus suggest that there may be a relationship between the istradefylline treatment and blood glucose increase. Indeed, blood glucose increase was among the AEs that occurred in Pool 1 with a higher frequency compared with placebo. Furthermore, in the FDA label, blood glucose increase is reported in the ADR table. Therefore, blood glucose increase was requested to be considered as an ADR with the frequency "uncommon" based on Pool 1 data.

ECG parameters

A thorough QTc study in healthy subjects showed no clinically relevant effects on QTc interval with 40 or 160 mg/day of istradefylline at steady-state

In the pooled safety data, there was no apparent differences in any of the mean values in the ECG parameters between the placebo and istradefylline treatment groups. Furthermore, a slightly higher proportion of subjects with treatment-emergent potentially clinically significant (PCS) ECG value was observed in the two proposed doses of istradefylline compared with placebo (20 mg 5.6% and 40 mg 4.4% vs. placebo 4.2%), in particular differences in QRS interval (3.5% and 4.1% vs. 2.0%), and QTcF (0.8% and 0.3% vs. 0%) were observed, and a higher proportion of subjects had treatment-emergent shifts in QRS interval in the istradefylline group compared to placebo (20 mg 2.4% and 40 mg 2.5% vs. placebo 1.1%). Similar findings for the QRS interval were recorded in Pool 2. Although the differences between istradefylline and placebo groups are not remarkably large, given the mechanism of action of istradefylline, the applicant was requested to discuss the potential for QT prolongation. Given the overall and specific ECG data provided by the applicant, particularly those from the QTc study, it appears that the effects of istradefylline on QT prolongation can be considered non-clinically significant.

Safety in special populations

Gender

There were no clear differences in the safety profile of istradefylline between females and males enrolled to studies. This is in line with the PK findings which showed only small differences in the exposure between genders.

Elderly

There was a trend towards slightly less favourable safety profile older subjects. The incidence of severe, serious TEAEs and TEAEs leading to discontinuation relative to the placebo group, was proportionately, greater for older subjects (≥ 65 years and ≥ 75 years) than for younger subjects. Dyskinesia and fall were proportionately more frequent in older ager groups (3% difference). In particular, relative to placebo, dyskinesia was proportionately more frequent for istradefylline-treated older subjects (≥ 65 years and ≥ 75 years) compared with istradefylline-treated younger subjects (< 65 years) (total istradefylline 17.8% and 12.8% vs. 17.8% placebo 8.5% and 6.1% vs. 10.6%). The applicant was requested to further discuss the safety in older subjects. As requested, the applicant analysed the safety data by age category. The largest age categories include subjects <65 years of age (46.5%, 1434/3083)

and 65 to 74 years of age (33.0%, 1017/3083). Fewer subjects were 75 to 84 (10.1%, 312/3083) and ≥ 85 (0.4%, 12/3083) years of age. It was concluded by the applicant that overall, the analysis of TEAEs by age groups did not reveal any concerning trends in older subjects. However, it is noted that in the 75- to 84-age category, subjects in the istradefylline 20 mg/day (21.7%) and 40 mg/day (17.8%) groups were reported to have higher frequencies of the grouped term "postural hypotension, falls, black outs, syncope, dizziness, ataxia, and fractures" compared to the placebo group (12.1%). The applicant considers however that the data do not suggest that istradefylline has less favourable safety profile in older subjects. Apart from dizziness, none of the AE listed in this grouped term are recognized as adverse drug reactions with istradefylline. In addition, it was highlighted that the size of the 75 to 84-year-old group is approximately 3 times smaller than that of 65 to 74-year-old group and approximately 4 times smaller than the < 65-year-old group, which may have resulted in more variance in the observed incidence of TEAEs in 75- to 84-year-old group. Limited data on the use of istradefylline exits in subjects' ≥85 years old.

Race and Region

AEs (both in placebo or istradefylline treatment groups) were more frequently reported in white patients and in North America as compared to Asian subjects and patients enrolled in other regions.

Otherwise, relative to the placebo group, the incidence of severe TEAEs and TEAEs leading to discontinuation was proportionally greater in Asian subjects as comparted to white subjects. This difference could be attributable to differences in the body weight.

The effect of the body weight on the exposure is not clear (please see the PK section for further details).

The applicant was requested to discuss the safety profile of istradefylline depending on the body weight. The relevant analysis was provided and it can be agreed with the applicant that no firm conclusion regarding differences in the safety profile depending on the BMI category can be made based on the available data.

A higher frequency of decreased appetite was observed in patients receiving treatment with istradefylline and decrease in body weight was observed in the majority of patients enrolled to long term studies.

The applicant has considered Decreased appetite and Weight decreased as ADRs with the frequency 'common' based on the incidence of these ADRs in Pool 1 data

Hepatic Impairment

The applicant performed two studies in patients with hepatic impairment. In study 6002-016 which investigated patients with mild impairment only of a single dose of 40 mg/day of istradefylline was given therefore this study has a limited value for safety assessment.

In a second study the treatment with 40 mg of istradefylline was given for 2 weeks.

In this study the highest percentage of AEs were reported in non-smokers with moderate hepatic impairment (85%) followed by non-smokers without hepatic impairments (42.9%) whereas in smokers irrespective of hepatic function less AEs were reported (less than 29%). This is could be explained by the PK profile of the drug which showed that the exposure is increased in patients with hepatic impairment but decreased in patients who smoke.

The types of AEs which were seen in this study were in line with those reported in pivotal studies.

As istradefylline was not studied in patients with severe hepatic impairment, use in severe hepatic impairment was considered as missing information in the RMP.

Renal impairment

There was no safety signal in the renal impairment study. Exposure is not significantly increased in patients with mild, moderate or severe renal impairment and therefore it is agreed that no dose adjustment is required in these patients. However, the product was not studied in patients end stage renal disease (CrCL <15 mL/min) or end stage renal disease requiring haemodialysis.

Interactions

Istradefylline is almost exclusively eliminated via metabolism. *In vitro* studies indicated that istradefylline was primarily metabolised by CYP1A1 and CYP3A4/5 with minor contribution of other CYPs. 7 DDI studies were performed by the applicant and these studies are discussed in the PK section of this AR.

The applicant analysed the Effect of Concomitant Treatments for PD.

Dyskinesia was reported more frequently in istradefylline-treated subjects who received concomitant Treatments for Parkinson's Disease with Dopamine agonists and COMT Inhibitors (19.0%, 22.6 % respectively) compared with istradefylline-treated subjects who did not receive these treatments concomitantly (13.8%, 14.4% respectively). There was no difference in the frequency of dyskinesia between istradefylline-treated subjects who received MAO-B inhibitors versus those without such treatment. Dyskinesia was also more frequent in patients concomitantly receiving 2 or 3 medications for Parkinson's Disease. SDyskinesia was reported more frequently in istradefylline-treated subjects who received DAs, MAO-B inhibitors, and COMT inhibitors (21.1%) compared with istradefylline-treated subjects who did not receive all 3 concomitant treatments (17.5%), although with a smaller difference compared with the concomitant treatment of 2 medications.

Based in the limited data it seems that there was trend towards a better safety profile of istradefylline in patients who smoked. This is in line with the PK finding which indicated the lower exposure in smokers.

The risk for the use in pregnancy

In non-clinical embryo-foetal development studies in rat fetotoxicity was evident at the high dose, limitations in the TK data in this study mean accurate derivation of exposure margins to predicted clinical exposures is impossible, but margins are likely < 3 fold. In rabbit, Istradefylline was teratogenic at high doses with the exposure margin to the NOAEL in this study = 2.5 fold that at predicted clinical exposures. Of note, in a rabbit embryo-foetal development study in which animals were administered Istradefylline with carbidopa and levodopa, teratogenicity was also observed. Exposure margins to the NOAEL in this study were lower = 1.2 fold i.e. within the range of anticipated clinical exposures (for more information see preclinical section of this AR).

Post-marketing data

Istradefylline was first approved with the brand name NOURIAST in Japan on 25 March 2013. Istradefylline was approved as NOURIANZ in the United States on 27 August 2019, at the same doses currently proposed (20 and 40 mg/day), for use as adjunctive treatment to L/C in adult patients with PD experiencing "OFF" episodes.

As of 31 May 2019, the cumulative post-marketing exposure estimate for istradefylline in Japan is approximately 63,500 patients.

The most frequently reported AEs in the post-marketing setting, dyskinesia, hallucinations, constipation, dizziness, nausea and vomiting are recognized as adverse reactions with istradefylline based on clinical trial data.

The most commonly reported serious AEs was pneumonia and pneumonia aspiration. Further discussion in relation to the frequency of pneumonia reported in the post-marketing setting was required. The applicant stated that it is not possible to calculate actual EAIRs for AEs of respiratory disorders reported in the post-marketing setting because these reports are mainly estimated from sales data and they do

not include detailed information on duration of exposure. Furthermore, it is acknowledged that post-marketing AEs are reported on voluntary basis and therefore could represent an underestimation of the actual incidence of AEs. Anyway, the applicant provided the reporting rate in the post-marketing setting calculated using the estimated exposure in patient-years as presented in the EU RMP SV.1 Post-authorisation exposure and compared them with EAIRs in the placebo group in Pool 1. These data indicate that post-marketing reporting rates for pneumonia and pneumonia aspiration were lower than EAIRs in the placebo group of Pool 1. It is acknowledged that none of the post-marketing respiratory AEs were reported with significantly higher rates than in the Pool 1 placebo group.

Serious hallucinations and other psychiatric disorders (delirium, delusion, depression and suicide attempt) have been reported also in the postmarking setting. As mentioned above, although these psychiatric symptoms are frequently present in patients with PD, there is some concern that istradefylline may increase frequency or increase their severity

2.6.2. Conclusions on the clinical safety

There are concerns that the higher proposed dose of istradefylline (40 mg/day) could be slightly less safe as compared to the lower dose especially in relation to the development of serious and severe TEAEs after a long term treatment with istradefylline.

As indicated in the efficacy sections of this assessment report, that there is no consistent evidence of greater efficacy with the 40mg compared to 20mg dose. For this reason, the justification for the use of the 40mg dose is being questioned.

2.7. Risk Management Plan

The CHMP and PRAC, having considered the data submitted in the application was of the opinion that due to the concerns identified with this application, the risk management plan cannot be agreed at this stage.

2.8. Pharmacovigilance

Pharmacovigilance system

The CHMP and PRAC, having considered the data submitted by the applicant, were of the opinion that, due to the concerns identified with this application, as above outlined, the pharmacovigilance system summary cannot be agreed at this stage.

Periodic Safety Update Reports submission requirements

Not applicable

2.9. New Active Substance

The CHMP, based on the available data, considers istradefylline to be a NAS as it is not a constituent of a medicinal product previously authorised within the European Union

2.10. Product information

In light of the negative recommendation, a satisfactory summary of product characteristics, labelling and package leaflet cannot be agreed at this stage.

2.10.1. User consultation

The results of the user consultation with target patient groups on the package leaflet submitted by the applicant show that the package leaflet meets the criteria for readability as set out in the *Guideline on the readability of the label and package leaflet of medicinal products for human use.*

2.10.2. Additional monitoring

Not applicable.

3. Benefit-Risk Balance

3.1. Therapeutic Context

3.1.1. Disease or condition

PD is a progressive, debilitating movement disorder characterised by bradykinesia, rigidity, and resting tremor that affects 1.2 million people in Europe (European Brain Council, 2019). Most motor symptoms of PD result from the progressive degeneration of the dopaminergic neurons in the substantia nigra pars compacta. As degeneration occurs, striatal concentrations of dopamine decrease, leading to reduced stimulation of dopamine receptors in the striatum. Dopamine activates the 'direct pathway' from the striatum of the basal ganglia via D1 receptors and suppresses the 'indirect pathway' from the striatum via D2 receptors. Thus, the reduced striatal dopamine that occurs in PD results in decreased activity of the direct pathway and increased activity of the indirect pathway. Increased excitability of the indirect pathway can be mediated by adenosine A_{2A} receptors in the striatum. The imbalance of activity between the direct and indirect pathways in PD is thought to result in the hallmark symptoms of bradykinesia, rigidity, tremor, and loss of postural reflexes.

3.1.2. Available therapies and unmet medical need

Levodopa remains the "gold standard" for therapy for PD that increases dopamine concentrations in the brain because levodopa can cross the blood-brain barrier and be transformed into dopamine. In the early stages of PD, patients usually experience substantial symptom relief from levodopa. Despite its initial efficacy, levodopa's therapeutic window narrows over time (Jankovic, 2005). That is, the duration of benefit from a dose of levodopa ("ON time") becomes shorter and PD symptoms return before the next scheduled dose ("wearing-off"). There are periods of time when, despite measurable plasma concentrations, levodopa does not control PD symptoms ("OFF time") and these periods become increasingly longer as PD progresses.

Efficacy of available treatments varies from patient-to-patient and over time. There are many different types of dopaminergic therapies that have been developed to try to treat OFF time (e.g., dopamine agonists, COMT inhibitors, MAO-B inhibitors); however, treatment is complicated by an increased risk of dyskinesia and a variety of side effects, including impulse control disorders and sleep disturbances, which may limit benefits or preclude continued use of these medications. Thus, despite available medical therapies, patients continue to suffer potentially disabling OFF episodes. Consequently, there is a continuing need for additional agents that are effective for the levodopa-treated patient (producing less OFF time and better symptom control) without intolerable side effects.

The clinical heterogeneity of PD signs and symptoms and in the individual levodopa requirements reinforces the need for additional adjunctive treatment options (Lewis, 2005).

Istradefylline (also known as KW-6002) is an adenosine A_{2A} receptor antagonist, which has a xanthine derivative structure and which competitively inhibits adenosine binding to the A_{2A} receptor. As an adenosine A_{2A} receptor it would represent an add-on treatment with a new mechanism of action for PD patients.

The target indication for istradefylline was as an adjunctive treatment to levodopa-based regimens in adult patients with PD experiencing "OFF" time.

3.1.3. Main clinical studies

The applicant has included 8 pivotal Phase 2b/3 randomized, double-blind, fixed-dose, placebo-controlled studies (6002-US-005, 6002-US-006, 6002-US-013, 6002-0608, 6002-009, 6002-US-018, 6002-EU-007, and 6002-014) and 2 open-label, long-term studies (6002-US-007, 6002-010). These studies in PD patients evaluated the safety and efficacy using 10, 20, 40, and 60 mg once daily doses of istradefylline.

Table 133: Study design comparison

6002-US-005	6002-US-006	6002-US-013	6002-0608	6002-009	6002-US-018	6002-EU-007	6002-014
Design: Double-blind, randomized, placebo-controlled, parallel-group clinical study							
Duration:							
12-week	12-week	12-week	12-week	12-week	12-week	16-week	12-week
Treatment Groups (randomization ratio):							
Istradefylline 40 mg/day or placebo (2:1 ratio)	Istradefylline 20 or 60 mg/day or placebo (2:2:1 ratio)	Istradefylline 20 mg/day or placebo (1:1 ratio)	Istradefylline 20 or 40 mg/day or placebo (1:1:1 ratio)	Istradefylline 20 or 40 mg/day or placebo (1:1:1 ratio)	Istradefylline 10, 20, or 40 mg/day or placebo (1:1:1:1 ratio)	Istradefylline 40 mg/day, or placebo, or entacapone (1:1:1 ratio)	Istradefylline 20 or 40 mg/day or placebo (1:1:1 ratio)
Subjects were: • At least 30 years of age (at least 20 years of age in Studies 6002-0608 and 6002-009); • Diagnosed with PD as determined by the UKPDS criteria; • Modified Hoehn and Yahr scale Stages 2 to 4 in the OFF state (Studies 6002-US-005, 6002-US-006, 6002-US-013, 6002-0608, 6002-009, 6002-US-018, and 6002-EU-007), and in the ON state (Study 6002-014); and • Had end-of-dose wearing-off with an average of at least 2 hours OFF time per day (Studies 6002-US-005, 6002-US-006, 6002-0608, 6002-009, and 6002-014) or 3 hours OFF time per day (Studies 6002-US-013, 6002-US-018, and 6002-EU-007) at study entry; subjects in 6002-014 also had levodopa-induced dyskinesia.							
Levodopa Requirements: • Receiving levodopa and a peripheral DOPA-decarboxylase inhibitor (carbidopa or benserazide) for at least 1 year. • Treated with levodopa for at least 1 year and have been on a stable regimen of levodopa for at least 4 weeks before randomization/baseline (as per protocol specification). – Studies 6002-US-005 and 6002-US-006: at least 4 doses/day, or at least 3 doses/day if 2 doses were slow-release formulations (no specific levodopa dose requirement). – Studies 6002-US-013, 6002-US-018, and 6002-EU-007 at least 3 doses/day (no specific levodopa dose requirement). – Studies 6002-0608 and 6002-009: at least 300 mg/day levodopa. – Study 6002-014 required subjects to be taking at least 400 mg/day levodopa plus at least 1 adjunctive dopaminergic medication approved to treat PD, and with documented levodopa-induced dyskinesia. • Decrease in the total daily dose of levodopa was permitted, if necessary (Investigator's discretion) due to levodopa-related AEs. • Change in either the frequency of levodopa dosing or the interval between levodopa doses was not allowed (Exception: Study 6002-EU-007 allowed levodopa dose adjustment during the initial 4 weeks).							

6002-US-005	6002-US-006	6002-US-013	6002-0608	6002-009	6002-US-018	6002-EU-007	6002-014
Other Parkinson's Medications were allowed (and were required in Study 6002-014). • Reduction in the dose of anti-parkinson medication was permitted to control dopaminergic-related AEs if a prior reduction in the levodopa dose was unsuccessful, or to alleviate AEs thought to be directly related to the agent being adjusted. • Prior anti-parkinson medication could not be increased and no new anti-parkinson medication could be added.							
Primary Endpoint (per protocol): Change from baseline in OFF time/day expressed as:							
% of awake time	% of awake time	% of awake time	Hours	Hours	% of awake time	% of awake time	Hours
Primary Statistical Methodology: Analysis Set[a]/Analysis Method (per protocol and SAP)							
ITT/ANOVA, ANCOVA[b] 12 week-LOCF, OC	ITT/ANOVA, ANCOVA[b] 12 week-LOCF, OC	ITT/ANCOVA 12 week-LOCF, OC	FAS/ANCOVA 12 week-LOCF, OC	FAS/ANCOVA 12 week-LOCF, OC	ITT/ANCOVA 12 week-LOCF, OC	ITT/ANCOVA 16 week-LOCF, OC	ITT/MMRM 12 week, OC

a the ITT and FAS analyses were essentially the same.
b The SAP prespecified ANOVA and the protocol prespecified ANCOVA
AE= Adverse event; ANCOVA=analysis of covariance; ANOVA=Analysis of variance; FAS=Full analysis set; ITT=intent-to-treat; LOCF=Last observation carried forward; MMRM=Mixed-effects model repeated measures; OC=Observed Case, PD=Parkinson Disease; SAP=Statistical analysis plan; UKPDS=United Kingdom Parkinson´s Disease Society.

3.2. Favourable effects

Four of the eight pivotal trials, met their primary endpoint showing a favourable reduction in the % change from baseline to endpoint in time spent OFF (studies US-005, US-013) or change from baseline to endpoint in total hours spent OFF (Japanese studies 6002-0608 and 6002-009).

In study US-005, for subjects in the istradefylline 40mg group, a greater mean decrease (-10.81%) from Baseline to Endpoint was observed in the percentage of awake time per day spent in an OFF state compared to that in the placebo group (-4.04%) and this difference was statistically significant (p-value = 0.007) based on the LSM from the ANOVA model. More meaningful clinically, change from baseline to endpoint in total hours spent in OFF time per day for istradefylline compared to placebo was performed

as a secondary endpoint without control for multiplicity thus results were solely descriptive. In this case, the difference between treatment groups of 1.15 hours less time spent in an OFF state for the istradefylline 40mg dose compared to placebo could be considered clinically relevant for patients.

In study US-013, at endpoint, the LSM reduction from Baseline in the percentage of awake time per day spent in the OFF state was 9.49% for the istradefylline 20mg group and 4.92% for the placebo group. This translated into a 4.57% greater reduction in the percentage of awake time per day spent in the OFF state for subjects in the istradefylline 20mg group compared to subjects in the placebo group; this reduction at Endpoint was statistically significant (p = 0.025) for the istradefylline 20mg group compared to the placebo group. In terms of LSM mean difference in total hours of awake time per day spent in OFF state, performed as a secondary efficacy endpoint analysis without control for multiplicity, this showed a reduction of 0.73 hours for istradefylline 20mg group compared to placebo. This approximates to a difference from placebo of 44 minutes.

Study 6002-009 (Phase 3) was essentially a replicate of Study 6002-0608. In study 6002-08. The LSM difference from placebo was -0.65 hours (95% C.I -1.23 to -0.07 hours) for the 20 mg/day KW-6002 group and -0.92 hours (-1.49 to -0.35 hours) for the 40 mg/day KW-6002 group. This approximates to a difference from placebo of 39 minutes for Istradefylline 20mg and 55 minutes for Istradefylline 40mg.

Study 6002-009 the LSM difference from the placebo group (95% CI) at the final evaluation was −0.76 h (−1.30 to −0.22 h) for the 20 mg/day of KW-6002 group and −0.74 h (−1.27 to −0.20 h) for the 40 mg/day of KW-6002 group. This approximates to a difference from placebo of approximately 45 minutes for both Istradefylline treatment arms.

No adjustment for multiplicity was performed for secondary endpoints in any of the pivotal studies other than study 6002-014, thus all secondary endpoints in general could only be considered descriptive. Considered most relevant clinically in terms of the secondary endpoints reported was change in ON time without troublesome dyskinesia (ON time without dyskinesia and ON time with non-troublesome dyskinesia). In general, for 6002-US-005, 6002-US-006, 6002-US-013 numerical increases suggesting potential for clinical improvement were seen for change in ON time without troublesome dyskinesia, even though statistically significant differences from PBO on this clinical important endpoint were observed only in study 6002-US-005, but not in study 6002-US-006 and 6002-US-013. However as aforementioned no control for multiplicity meant these could only be considered descriptive.

3.3. Uncertainties and limitations about favourable effects

Study US-006 was a formally failed study as per the prespecified primary analysis model ANOVA. However, the primary analysis model was changed from ANOVA to ANCOVA after finalising the SAP and even with changing the analysis model, the primary endpoint result for the overall treatment effect was marginal for significance with p=0.049.

Study 6002-US-018 did not meet its primary endpoint and in fact, this study demonstrated numerical worsening for istradefylline 10mg and 20mg compared to placebo with only a very minor numerical improvement for istradefylline 40mg compared to placebo that was not statistically significant nor clinically meaningful with a reduction in OFF time compared to placebo of 0.03 hours for istradefylline 40mg. For the secondary endpoint, ON time without troublesome dyskinesia, in study 6002-US-018 for all istradefylline doses 10, 20 and 40mg there was a numerical decrease found which could be considered clinically unfavourable. None of the secondary endpoints were controlled for multiplicity thus this was only considered descriptive.

Study 6002-EU-007 did not meet its primary endpoint Istradefylline 40mg failed to show any benefit in the only pivotal study conducted with an active comparator (entacapone 200mg daily) as well as a placebo. In fact, there was no difference between Istradefylline 40mg in percentage reduction of OFF

from baseline at endpoint (-4.53% for placebo and -5.14% for Istradefylline 40mg) and placebo. The reduction from baseline for entacapone was slightly greater at -7.82%. The difference versus placebo was not statistically significant.

Neither was any efficacy demonstrated in the most recent study, (Study 6002-014) which aimed to enrol a 'maximally and optimally treated' PD population, who also had a history of levodopa induced dyskinesia, and were on higher baseline doses of levodopa (mean dosage in excess of 800mg), and on a range of adjunctive treatments, i.e at least 1 other dopaminergic antiparkinson medication. The fact that there is such a degree of inconsistency in study results adds considerable uncertainty to the overall assessment of this marketing authorisation application.

The two positive US studies (US-005 and US-013) were conducted between 2002-2003, over 15 years ago, making their generalisability to current treatment settings questionable.

The more recently conducted trials including study US-014 completed in 2016, did not demonstrate any efficacy, however it is notable this was in a distinct, presumably more severe population with presence of dyskinesias and requiring >/=400mg levodopa, plus a clinically effective dose of at least 1 other dopaminergic antiparkinson medication.

There remains considerable uncertainty as to how any of the positive efficacy findings could apply to a European population, considering both studies conducted with European subjects (6002-EU-007 and US-014) were negative for demonstrating efficacy.

It remains unclear whether there is any impact of race/ethnicity on efficacy of istradefylline treatment. However, it is noted that more favourable results were seen in both Japanese studies.

Given the inconsistency of the individual study results the applicant undertook a pooled analysis of the 8 pivotal studies. This pooled analysis had not been predefined, but was performed as a rescue measure following the individual trials that were planned to stand on their own but showed only borderline or no effect. Initially a fixed effects model and later - given the heterogeneity of the studies considering their regional and wide temporal spread - a more appropriate random effects model was used. Importantly, by the nature of these *post hoc* defined analyses, these analyses could only be performed on an exploratory level without adjustments for multiplicity. The estimated effect size with the random effects meta-analysis was modest and while the 95%-CIs excluded '0', this nominal significance level can formally not be used to establish statistical significance. Against this background, and given the proximity of the CI-boundaries to 'no effect' lack of a treatment effect cannot be excluded. Besides this a larger sample size (as a consequence of meta-analytic pooling of different studies) allows smaller p-values in case of small effects, which would also not be convincing. The reduction in OFF time of 0.45 hours (95% CI:-0.75, -0.15 hours) for Istradefylline 20mg and -0.46 hours (95% CI: -0.8, -0.12 hours) for Istradefylline 40mg, corresponds to a decrease in daily OFF time of only 27 minutes. Likewise the increases in ON time without troublesome dyskinesia were small, 0.43 hours (95% CI: 0.14, 0.73) for Istradefylline 20mg and 0.34 hours (95% CI -0.01, 0.69 hours) corresponding to an improvement in ON time without troublesome dyskinesia of 26 and 20 minutes respectively.

There has also been no clear rationale for the dosing strategy of both 20mg and 40mg with no clear instruction for prescribers as to when to use a particular dose. In the 4 trials that studied both the 20 mg/ day and the 40 mg/ day dose, discordant results were observed. A trend towards a greater effect in reduction of OFF time and increase in ON time without troublesome dyskinesia for 40 mg vs 20 mg/ day was observed in study 018 (that was a negative study, showing no difference vs PBO) and in Study 0608 (Japanese study). Conversely, no difference between 20 and 40 mg/ day dose was observed in reduction of OFF time and increase in ON time without troublesome dyskinesia, in the other Japanese study (0609) and in Study 014.

Furthermore, the pooled results for 8 RCTs did not show relevant differences between the 20 and the 40 mg dose in total OFF (-0.38hr vs -0.45hr) nor in ON with non-troublesome dyskinesia (0.25hr for both doses). This further increases the overall uncertainty with regard to the observed efficacy and lack of efficacy in the different pivotal studies.

3.4. Unfavourable effects

TEAEs were reported more frequently in patients receiving istradefylline treatment. In Pool 1, 72.4% of subjects in the total istradefylline group experienced at least 1 TEAE compared to 65.4% of subjects in the placebo group. In Pool 2 89.7% of subjects reported any TEAEs.

Dyskinesia was the most commonly reported TEAE. In Pool 1 the incidence of dyskinesia was greater in the total istradefylline group (17.8%) compared with the placebo group (9.6%), with similar incidences of dyskinesia in the istradefylline 20 mg/day (16.1%) and istradefylline 40 mg/day (17.7%) groups. For dyskinesia the difference between the istradefylline group and placebo was 8%. In addition, dyskinesia was the most frequently reported severe TEAEs and the most frequently reported TEAE leading to discontinuation although the number of these patients was small.

For other TEAEs reported with the higher frequency in the istradefylline group the difference as compared to placebo groups was smaller i.e for nausea there was 2% differences, for dizziness 1 % difference, constipation 1.6 % difference and hallucination 1% difference.

The number of patients who experienced serious TEAEs in Pool 1 was small however, the percentage of such patients was slightly higher in the istradefylline group (4.2%) as compared to the placebo group (3.1%). More serious TEAEs were reported in patients receiving 40 mg dose as compared to those treated with 20 mg. The most frequently reported serious TEAEs were falls and pneumonia and for those there was no significant differences between istradefylline and placebo. Cardiac failure congestive and delirium were reported in 3 patients treated with istradefylline versus 0 cases in the placebo group.

Nine subjects died in Pool 1 and thirty-two subjects died in Pool 2 however, the provided data do not indicate that there is a higher risk of death of patients receiving treatment with istradefylline.

In Pool 1 TEAEs within the SOC Psychiatric disorders were reported more frequently in the istradefylline group (16.2%) as compared to those receiving placebo (11.4%). The highest frequency was seen in the highest dose group (24.5% in patients receiving 60 mg dose). TEAEs Anxiety had a slightly higher frequency in Pool 1 and in Pool 3.

Also serious TEAEs within the SOC Psychiatric disorders were reported with the higher frequency in the istradefylline group (10 cases in total) as compared to 1 case in the placebo group. The following serious TEAEs were reported: confusion sate, delirium, psychotic disorders, depression, disorientation, hallucination, persecutory delusion, psychiatric symptoms, suicidal attempt. It is noted that patients with psychotic illness or depression were excluded from all studies. Psychotic disorders including mania, agitation, delirium, and abnormal behaviour, with some of these that considered serious, were reported only in patients treated with the proposed doses of istradefylline versus no case in the placebo group. Serious TEAEs of anxiety, confusional state, hallucination, persecutory delusion was also reported in patients receiving istradefylline in Pool 3 and also in this pool no serious TEAEs within the SOC Psychiatric disorders were reported in the placebo group. The AE hallucinations was originally proposed by the applicant to be listed as ADR.

Psychotic disorders such Hallucination, Insomnia, Vivid dreams, Paranoia, Delusions, Mania, Confusional state, Abnormal behavior and Agitationas are included as ADR Table with the frequency "uncommon" based on the Pool 1 data.

The slightly higher frequency of the upper respiratory tract infections observed in Pool 1 is apparently mainly driven by upper respiratory tract inflammation term. Respiratory TEAEs were the most frequently reported also in the long-term open-label Pool 2, and both upper and viral upper respiratory infections occurred with a higher incidence also with istradefylline monotherapy compared to placebo. Given the above and the biological plausibility related to the istradefylline mechanism of action, as requested, the applicant has added upper respiratory tract inflammation as an ADRs.

The higher frequency of decreased appetite was observed in patients receiving treatment with istradefylline and that decrease in body weight was observed in the majority of patients enrolled to long-term studies.

In controlled studies (Pool 1), adverse reactions consistent with impulse control disorder (including hypersexuality, hyperphagia, impulse control, increased libido, gambling, increased appetite, and obsessive-compulsive thoughts/disorder) were observed in 0% of patients in the placebo group, and in 0.7%, 0.8%, 0.6%, and 0% of patients in the istradefylline 10, 20, 40, and 60 mg/day groups, respectively. Although the differences in incidence between istradefylline and placebo were small, all cases of impulse control disorder (n=13) occurred in istradefylline-treated subjects. No serious ICD was reported, and discontinuation for ICD was observed only in one patient. Overall, 11 out of 13 subjects who experienced ICDs were on concomitant treatment with DAs. In the open-label long-term Pool 2, the majority of subjects with ICD were on concomitant DA treatment and resolved while istradefylline was continued.

Although sleep disorders are common in patients with PD, insomnia and abnormal dreams were reported more frequently in the istradefylline group compared with placebo. Insomnia also resulted one of the most commonly reported AEs in the long-term Pool 2. Insomnia and Vivid dreams are now included in the ADR Table of section 4.8 with the frequency "uncommon" based on the Pool 1 data.

Blood glucose increase was among the AEs that occurred in Pool 1 with a higher frequency compared with placebo. Furthermore, in the FDA label, blood glucose increase is reported in the ADR table. Therefore, blood glucose increase was requested to be considered as an ADR.

Istradefylline is almost exclusively eliminated via metabolism. *In vitro* studies indicated that istradefylline was primarily metabolised by CYP1A1 and CYP3A4/5 with minor contribution of other CYPs.

In non-clinical embryo-foetal development studies in rat fetotoxicity was evident at the high dose, limitations in the TK data in this study mean accurate derivation of exposure margins to predicted clinical exposures is impossible, but margins are likely < 3 fold. In rabbit, Istradefylline was teratogenic at high doses with the exposure margin to the NOAEL in this study = 2.5 fold that at predicted clinical exposures. Of note, in a rabbit embryo-foetal development study in which animals were administered Istradefylline with carbidopa and levodopa, teratogenicity was also observed. Exposure margins to the NOAEL in this study were lower = 1.2 fold i.e. within the range of anticipated clinical exposures.

As of 31 May 2019, the cumulative post-marketing exposure estimate for istradefylline in Japan is approximately 63,500 patients.

The most frequently reported AEs in the post-marketing setting, dyskinesia, hallucinations, constipation, dizziness, nausea and vomiting are recognized as adverse reactions with istradefylline based on clinical trial data.

The most commonly reported SAEs were pneumonia and pneumonia aspiration.

During routine signal detection activities, a safety signal of rash with istradefylline was detected from post-marketing data in Japan.

3.5. Uncertainties and limitations about unfavourable effects

There are concerns that the higher proposed dose of istradefylline (40 mg/day) could be slightly less safe as compared to the lower dose especially in relation to the development of serious and severe TEAEs after a long-term treatment with istradefylline.

In Pool 2 serious TEAEs were reported in 16.1% of subjects receiving 20 mg dose as compared to 25.9% subjects receiving the 40 mg dose. The incidence per patient-year exposure of serious TEAEs was 0.27 in the 20 mg/day group and 0.34 in the 40 mg/day group in Pool 2.

Severe TEAE were also reported more frequently (in 28.3%) in the 40 mg treatment group as compared to 19.5% in the 20 mg treatment group and the difference between the treatment groups was particularly seen in relation to severe TEAEs considered to be ADR, e.g., hallucinations, nausea and vomiting. In long term studies, the overall EAIR of severe TEAEs was numerically higher in the 40 mg/day group (0.46) as compared to the 20 mg/day group (0.39).

Based on the presented results it could be concluded that in respect to serious, severe TEAEs and ADRs the differences in EAIRs between the 20 mg/day group and the 40 mg/day group were small. Nevertheless, it seems that during a long-term treatment the 40 mg dose could be associated with a slightly less favourable safety profile as serious and severe TEAEs were reported more frequently in patients receiving higher dose.

There are still some uncertainties in relation to the development of foci of vascular mineralization which was seen in preclinical studies.

The applicant claims that the available safety data provide reassurance that this concern is not relevant to humans. This is not fully agreed. The safety data from Pool 1 are considered to be of a limited value in the context of this safety concern, as 3 months exposure to istradefylline in Pool 1 is likely to be too short for the development of these brain changes. The review of the Pool 2 or postmarking data is considered to be more relevant however, there are some limitations. Possible symptoms of brain vascular mineralisation are likely to be difficult to be distinguished from symptoms of PD or AEs linked to PD medications. The applicant examined the post-marketing safety database for findings that might suggest development of foci of vascular mineralization in the brains of human patients was provided. However, this review was performed 5 years after authorisation in Japan and it is not clear if this exposure to istradefylline was long enough. The applicant agreed to add brain vascular mineralisation to the RMP as an important potential risk subsequently re-named as 'Movement, neurological or psychiatric disorders due to brain vascular mineralisation'. It was considered however that this important potential risk could be further re-named as follows: "Neurological (mainly movement) disorders or psychiatric disorders which the applicant agreed on.

A further discussion was required in relation to the potential risk of cardiac disorders taking into consideration the mechanism of action of istradefylline, which through its antagonistic action on the A_{2A} receptors could mediate coronary artery vasoconstriction, increase heart rate and blood pressure. Cases of cardiac SAEs were presented by the applicant which were considered as possibly related to istradefylline. Taking into account the biological plausibility related to the istradefylline mechanism of action and differences in the frequency of TEAEs compared with placebo reported in studies, there are concerns that the use of istradefylline could be associated with an increased risk of upper respiratory tract inflammation or other respiratory disorders. Worsening of pre-existing respiratory conditions such as asthma or chronic obstructive pulmonary disease may occur. The applicant have thoroughly discussed the cardiovascular AE-related issue based on both non-clinical and clinical available data. Given the above, the applicant concluded that these data do not support a clear direct relationship of istradefylline treatment with cardiac eventsThe applicant was requested to commit monitoring of any signal on cardiac

events suggestive of potential causal relationship with istradefylline treatment in the post-marketing setting.

The safety data on istradefylline for patients over 85 years of age are very limited and, hence, definitive conclusions on safety in this subpopulation cannot be made.

3.6. Effects Table

Table 134: Effects Table for Istradefylline 20mg and 40mg as adjunctive treatment to levodopa

Effect	Short Description	Unit	Treatment	Control	Uncertainties/ Strength of evidence	References
	Favourable Effects					
Positive Studies			**Placebo**	**Istradefylline 20mg**	**Istradefylline 40mg**	
Study US-005 Reduction in OFF Time	Primary endpoint % change from baseline to endpoint in awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	-3.71%	N/A	-10.49% -6.78% -11.63, -1.92 0.007	Same effect size not replicated consistently in other studies Confidence intervals are wide Conducted only in North America, >15 years ago	
Study US-013 Reduction in OFF Time	Primary endpoint % change from baseline to endpoint in awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	-4.92%	-9.49% -4.57% -8.55, -0.59 0.025	N/A	Conducted only in North America, >15 years ago Confidence intervals are wide	
Study 6002-0608	Primary endpoint Change from baseline to endpoint in HOURS awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	-0.66h	-1.31h -0.65h -1.23, -0.07 0.028	-1.58h -0.92h -1.49, -0.35 0.002	Conducted in Japan, greater proportion of female participants Same effect size not replicated consistently in other studies	
Study 6002-009	Primary endpoint Change from baseline to endpoint in HOURS awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	-0.23h	-0.99h -0.76h -1.30, -0.22 0.006	-0.96h -0.74h -1.27, -0.20 0.008	Conducted in Japan, greater proportion of female participants Same effect size not replicated consistently in other studies No difference between the 20mg and 40mg doses	

Effect	Short Description	Unit	Treatment	Control	Uncertainties/ Strength of evidence	References
Negative Studies (included considering there were pivotal studies)			**Placebo**	**Istradefylline 20mg**	**Istradefylline 40mg /(60mg)***	
Study US-006 Reduction in OFF Time	Primary endpoint % change from baseline to endpoint in awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	-4.07% ANOVA	20mg -7.72% -3.65% -7.83, 0.53 0.088	60mg -7.84% -3.77% -8.01, 0.47 0.082	Prespecified analysis was ANOVA, however also performed ANCOVA which was marginal for statistical significance No difference between 20mg and 60mg doses Conducted only in North America, >15 years ago	
Study US-018	Primary endpoint % change from baseline to endpoint in awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	-8.31%	-6.81% 1.50% -2.05,5.05 0.408	-8.97% -0.66% -4.21, 2.88 0.714	Statistical significance not reached. Istradefylline 10mg dose also included. Istradefylline 10mg and 20mg were both inferior to placebo	
Study EU-007	Primary endpoint % change from baseline to endpoint in awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	Placebo -4.53%	**Entacapone 200mg** -5.14% -3.29% -6.77, 0.19 0.064	Istra 40mg -7.82% -0.61% -4.05, 2.83 0.729	Did not meet statistical significance for Istra 40mg in European population and numerically Entacapone active comparator performed better, also when comparing CI.	
Study US-014	Primary endpoint Change from baseline to endpoint in HOURS awake time spent in OFF state MMRM	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	Placebo -0.88 h	Istra 20mg -1.20 h -0.32 h -0.76, 0.12 0.156	Istra 40mg -1.15 h -0.27 h -0.70, 0.17 0.234	Failed to show a benefit in a more 'severe' population required to have dyskinesias and be on >/=400mg levodopa/day No difference between 20mg and 40mg doses	

Effect	Short Description	Unit	Treatment		Control	Uncertainties/ Strength of evidence	References
Pooled Analysis of 8 RCTs Pool E1	Total HOURS OFF	LSM ean change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	Placebo	Istra 20mg -0.38 -0.61, -0.15	Istra 40mg -0.45 -0.68, -0.22	Effect size not compelling and clinical relevance is questioned for OFF time and ON time without troublesome dyskinesias No evident difference in treatment effect between 20mg and 40mg istradefylline doses	Table 2.7.3-17 SCE
	Total HOURS ON without troublesome dyskinesia	LSM difference placebo in change from baseline to endpoint 95% CI p-value		0.40 0.15, 0.66	0.33 0.08, 0.59		
Unfavourable Effects							
	Incidence of Dyskinesia %	%	17.2		9.2		
	Incidence of Nausea	%	5.9		3.7		
	Incidence of Dizziness	%	4.4		3.4		
	Incidence of Constipation	%	4.1		2.5		

Notes: Only primary endpoint information is included in this table considering additional secondary endpoints were not controlled for multiplicity.

3.7. Benefit-risk assessment and discussion

3.7.1. Importance of favourable and unfavourable effects

Considering the overall inconsistency in study results, istradefylline's clinical effect cannot be determined with any certainty.

Only four out of eight randomised DB placebo-controlled trials demonstrated efficacy in meeting the primary endpoint of reduction in time spent in OFF state (% for 6002-US-005 and 6002-US-013 and total hours for 6002-0608 and 6002-009). Moreover, treatment-induced reduction in the time spent in the OFF state is inconsistently accompanied by statistically significant increases in the ON state without troublesome dyskinesia, across the 4 positive trials.

The only two clinical studies that succeeded in showing a concurrent improvement with istradefylline treatment on both OFF and ON states, are methodologically weak as the selection of the ON endpoint was done post hoc or with no adjustment for multiplicity. Further limitations of the evidence supporting treatment benefit, come from the lack of confirmation of treatment efficacy in improving motor function, (by UPDRS III score), and ameliorating clinical global status of patients (as per CGI scale) in the two positive trials in Caucasian patients.

Such reductions in OFF time must be considered in the context of three further pivotal trials where the efficacy endpoint was not reached and one trial in which there was unjustified changes made to the statistical analysis thus yielding a very marginally positive result.

It also needs to be borne in mind that the positive US studies were conducted over 15 years ago and given the availability of newer therapies, the study results may not be generalisable to a present day European population, especially considering the two studies which enrolled European patients were negative studies, 6002-EU-007 and 6002-014.

A post hoc random effects meta-analysis of the eight pivotal studies investigating change from baseline in OFF time and ON time without dyskinesia compared to placebo for the Istradefylline 20mg and 40mg dose has showed modest estimated effect sizes, while the 95%-CI excluded '0', this nominal significance level can formally not be used to establish statistical significance. Against this background, and given the proximity of the CI-boundaries to 'no effect' a lack of a treatment effect cannot be excluded. Besides this a larger sample size (as a consequence of meta-analytic pooling of different studies) allows smaller p-values in case of small effects, which would also not be convincing. The clinical relevance of the estimated effect size is also questioned.

There has also been no clear rationale for the dosing strategy of both 20mg and 40mg. This further increases the overall uncertainty with regard to the observed efficacy and lack of efficacy in the different pivotal studies. Overall, there is no evidence from the efficacy studies to support greater efficacy for the 40mg Istradefylline dose compared to the 20mg dose. There are concerns that the higher proposed dose of istradefylline (40 mg/day) could be slightly less safe as compared to the lower dose especially in relation to the development of serious and severe TEAEs after a long-term treatment with istradefylline.

In Pool 2 serious TEAEs were reported in 16.1% of subjects receiving 20 mg dose as compared to 25.9% subjects receiving the 40 mg dose. The incidence per patient-year exposure of serious TEAEs was 0.27 in the 20 mg/day group and 0.34 in the 40 mg/day group in Pool 2.

Severe TEAE were also reported more frequently (in 28.3%) in the 40 mg treatment group as compared to 19.5% in the 20 mg treatment group and the difference between the treatment groups was particularly seen in relation to severe TEAEs considered to be adverse drug reactions, e.g., hallucinations, nausea and vomiting. In long term studies, the overall EAIR of severe TEAEs was numerically higher in the 40 mg/day group (0.46) as compared to the 20 mg/day group (0.39).

Based on the presented results it could be concluded that in respect to serious, severe TEAEs and ADRs the differences in EAIRs between the 20 mg/day group and the 40 mg/day group were small. Nevertheless, it seems that during a long-term treatment the 40 mg dose could be associated with a slightly less favourable safety profile as serious and severe TEAEs were reported more frequently in patients receiving higher dose.

3.7.2. Balance of benefits and risks

Despite four positive trials that met the primary efficacy endpoint, there are four formally negative trials where clinical benefit of istradefylline has clearly not been shown (one of which may provide marginal support for benefit using an additional analysis (6002-US-006). The degree of inconsistency despite similarly designed clinical studies raises concerns over the potential for this drug to provide benefit to patients.

A *post hoc* random effects meta-analysis of the eight pivotal studies investigating change from baseline in OFF time and ON tine without dyskinesia compared to placebo for the Istradefylline 20mg and 40mg dose has shown modest estimated effect sizes, with wide CIs for both doses. Whilst the 95%-CI excluded '0', this nominal significance level can formally not be used to establish statistical significance. Against

this background, and given the proximity of the CI-boundaries to 'no effect' a lack of a treatment effect cannot be excluded.

Importantly, half of the studies including those enrolling European patients and the most recent trial which enrolled a theoretically more severe PD population have not demonstrated any evidence of clinical benefit with addition of istradefylline to ongoing PD treatment. Even for the studies in which some efficacy has been shown, both Japanese studies, the higher proportion of females makes generalisation to European population difficult, and the clinical relevance of any observed treatment effect size is questioned.

There has also been no clear rationale for the dosing strategy of both 20mg and 40mg. The studies did not demonstrate any additional benefit for the 40mg over the 20mg Istradefylline dose, and no dose dependency has been shown between the doses.

The applicant was invited to attend an oral explanation at the meeting of the CHMP on June 23rd, 2021 to address the remaining major objections, in particular as to whether efficacy had been demonstrated. The arguments presented by the applicant were considered by the CHMP (see clinicial discussion).

In conclusion, the CHMP concluded following the oral explanation and discussion that efficacy had not been demonstrated and that the benefit risk balance for Istradefylline was negative.

3.8. Conclusions

The overall B/R of Nouryant is negative.

4. Recommendations

Outcome

Based on the CHMP review of data on quality, safety and efficacy for Nouryant in the treatment of adults as an adjunctive treatment to levodopa based regimens in patients with Parkinson's disease (PD) experiencing "OFF" time, the CHMP considers by consensus that the efficacy of the above mentioned medicinal product is not sufficiently demonstrated, and, therefore recommends the refusal of the granting of the marketing authorisation for the above mentioned medicinal product. The CHMP considers that:

The applicant submitted eight pivotal clinical studies in support of this application. While four of the eight pivotal trials submitted met their primary endpoint of a reduction in the % change from baseline to endpoint in time spent OFF (studies US-005, US-013) or change from baseline to endpoint in total hours spent OFF (Japanese studies 0608 and 009), the other trials did not.

Therefore, the efficacy of the product is not demonstrated considering the inconsistency of the results across the development program. More specifically:

- Study US-006 was a formally failed study as per the pre-specified primary ANOVA analysis model. The primary analysis model was changed from ANOVA to ANCOVA after finalising the SAP and even with changing the analysis model the primary endpoint result was marginal, achieving a nominal p-value of p=0.049. Considering the late change in the statistical analysis, this study is not considered to have demonstrated a statistically significant treatment effect.
- The inconsistency of study results including the different responses observed in the studies in different regions and the difference in response over time, are not resolved by post-hoc pooling

the individual trials intended to rescue their unconvincing results. In addition, the estimated modest effect size, and the unadjusted nominal confidence interval that is in close proximity to no effect are not considered sufficient to have demonstrated a treatment effect in a post-hoc pooled analysis. Moreover the observation of no clear pattern of a dose response with increasing doses of istradefylline, leads to considerable uncertainty on the effect of the treatment.

- The fact that Studies 007 and 014 (that enrolled the European population) both clearly failed remains an unresolved issue. In particular, as the most recent study, Study 6002-014 in 'maximally and optimally treated' patients did not show positive results – despite being the largest pivotal trial relative to size of each treatment group and despite being planned, taking into account the results of previously conducted istradefylline's trials.

The CHMP is of the opinion that the efficacy of the above-mentioned medicinal product is not properly, or sufficiently demonstrated. Therefore, the CHMP has recommended the refusal of the granting of the marketing authorisation for Nouryant.

Furthermore, following review of the available data in the context of the applicant's claim of new active substance status, the CHMP position at the time of this report is reflected in section 2.9 (new active substance). However, in light of the negative recommendation, the CHMP is of the opinion that it is not appropriate to conclude on the new active substance status at this time.

5. Re-examination of the CHMP opinion of 22 July 2021

Following the CHMP conclusion that Nouryant was not approvable based on the grounds for negative opinion described in section 4 above, the applicant submitted detailed grounds for the reexamination of the grounds for refusal.

5.1. Detailed grounds for re-examination submitted by the applicant

Following a request from the applicant at the time of the re-examination, the CHMP convened a Scientific Advisory Group (SAG) inviting the experts to provide their views on the CHMP grounds for refusal, taking into account the applicant's response detailed below:

Of note, in their response to the Grounds for Refusal, the applicant uses a different ordering of the ground for re-examination compared to the grounds for refusal. The following assessment will follow the applicant's ordering but with the original ordering added to the headings.

5.1.1. Ground #1 (the introduction plus dot 2 of the CHMPs GfR)

Grounds for re-examination:

The applicant disagrees with the CHMP conclusion that the efficacy of istradefylline has not been demonstrated and asserts that the consistency, certainty, and magnitude of the effect of istradefylline have been established and that due to similarity of the study designs, pooling of data is appropriate to support efficacy of istradefylline.

applicant's Position on Ground 1

- *The certainty of the treatment effect of istradefylline has been clearly demonstrated by numerically positive results in 7 of the 8 pivotal studies for the primary endpoint, 4 of which were formally statistically significant, whilst a fifth study met the primary endpoint when the most appropriate statistical method of analysis, pre-specified in the study protocol, is applied.*

- *The observed treatment effects across the pivotal study program are consistent and fall within the predicted range that takes into account the inherent heterogeneity of PD.*
- *Appropriately conducted random effects meta-analyses confirm the certainty and magnitude of effect of istradefylline when all pivotal studies were assessed.*
- *Baseline OFF time hours was found to be the main treatment effect modifier. The treatment effect increases in magnitude as baseline OFF time increases providing further evidence of the certainty of effect of istradefylline.*

Introduction

This ground is structured to demonstrate the certainty and consistency of the effect of istradefylline. Section "Certainty of Effect" provides the main evidence for demonstrating the certainty based on the individual study results, and this is further evidenced by random effects meta-analyses on the istradefylline 20 mg/day and 40 mg/day doses separately and combined, along with additional supportive analyses. Section "Consistency of Effect" demonstrates the consistency of the observed treatment effect across the pivotal study program. Section "Justification of Retrospective Meta-Analysis" presents a justification of the use of retrospective meta-analyses which strongly support conclusions of efficacy.

The istradefylline program included 8 pivotal studies as detailed in Table 133. The amount of awake time spent in the OFF state was the basis of the primary endpoint in each of the studies; change from baseline in hours OFF time was the pre-specified primary endpoint in Studies 6002-0608, 6002-009 and 6002-014, while the change from baseline in percentage of awake time spent in the OFF state was the primary endpoint in Studies 6002-US-005, 6002-US-006, 6002-US-013, 6002-US-018, and 6002-EU-007.

The primary efficacy variable in the MAA SAP was the change from baseline in total hours per day spent in the OFF state for the pooled analyses. This variable was used subsequently in various additional analyses that combined data across the pivotal study program.

The key secondary variable in the MAA Statistical Analysis Plan was the amount of time spent in the ON state without troublesome dyskinesia while the UPDRS Part III score measured in the 'ON' state was a further secondary endpoint of interest as it is a motor evaluation of disability in PD.

Discussion in this Ground will focus on the evidence provided by these 3 variables for certainty of effect, the certainty of the magnitude of effect, and the consistency of effect.

This Ground will use data from all 8 studies (Pool E1). Four studies randomized subjects to either istradefylline 20 mg/day, istradefylline 40 mg/day or placebo (Pool E4)[1]; these studies allow a comparison of the 2 dose levels while respecting the randomization. These data will be used in some discussions regarding the presence of a dose dependent effect that further supports the certainty of effect.

Note that all p-values are 2-sided with statistical significance judged at the 5% level.

Certainty of Effect

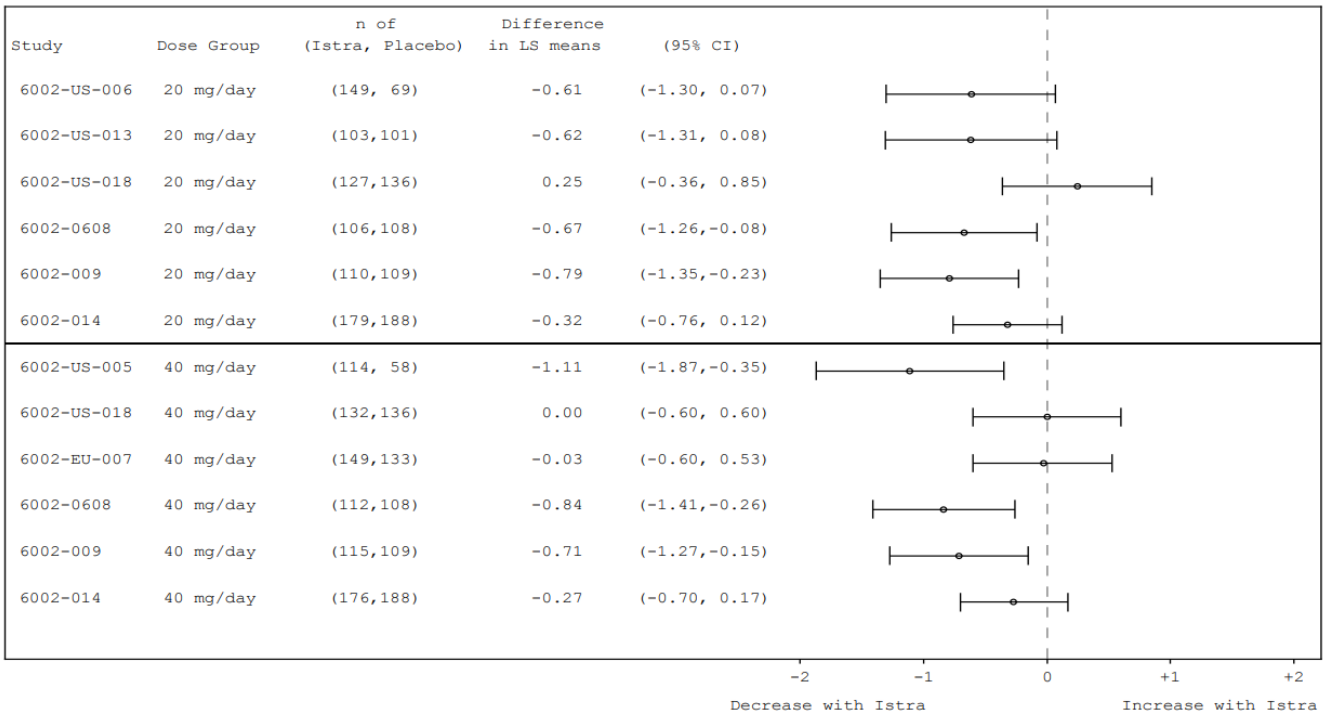
Main Evidence for Demonstrating Certainty of Effect

Study-level data for the change from baseline in OFF time to Week 12 for 20 mg/day and 40 mg/day of istradefylline compared to placebo is presented in Figure 44. Note that each study was of 12 weeks' duration, with the exception of Study 6002-EU-007, which was of 16 weeks' duration. The forest plots presented in this Ground focus on the 12-week outcomes (including Study 6002-EU-007) in order to

[1] Study 6002-US-018 also randomized subjects to istradefylline 10 mg/day.

provide a common endpoint across the program for the purposes of consistency. The Week 16 data for Study 6002-EU-007 were comparable to the Week 12 data.

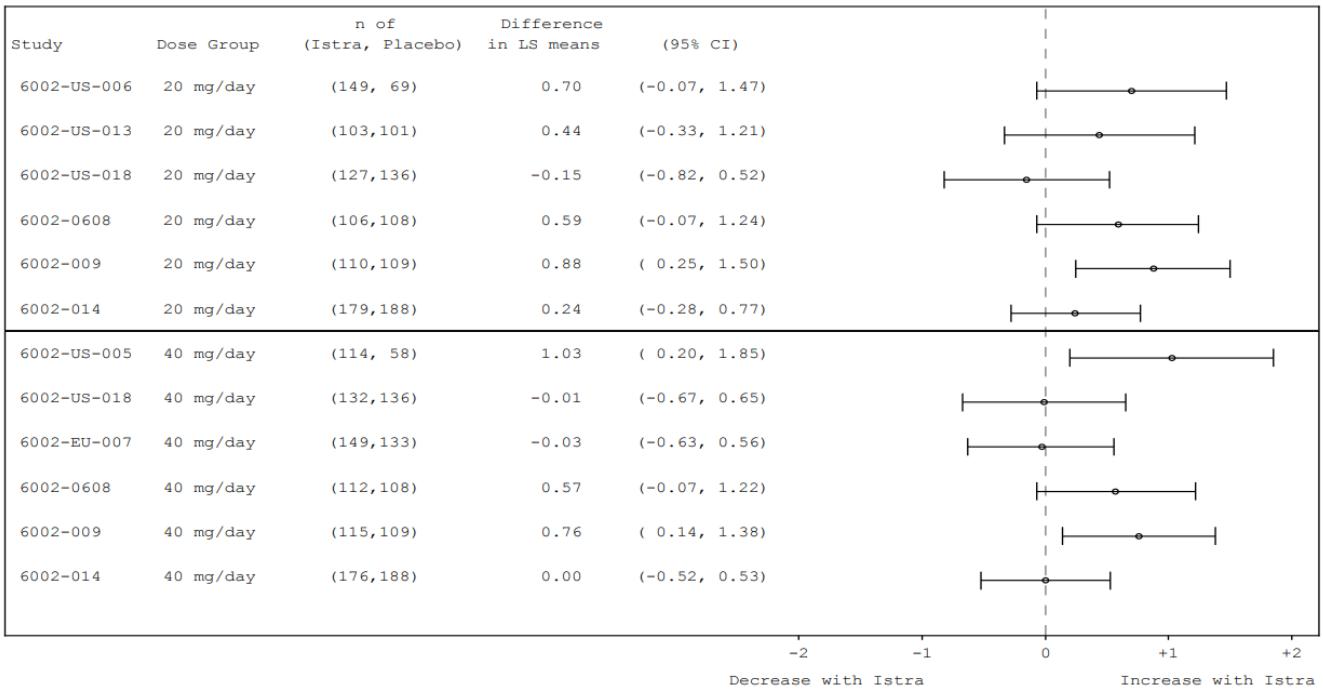
Figure 44: LS Mean Difference from Placebo in the Change from Baseline to Week 12 hours/day in the OFF State [95%CI] for Istradefylline (20 mg/day and 40 mg/day) - (OC, ITT, MMRM)



Source: Module 5, Re-exam Figure 9.1.

There are 12 comparisons of istradefylline against placebo for the change from baseline in OFF time: 6 for istradefylline 20 mg/day and 6 for istradefylline 40 mg/day. The majority of these comparisons are numerically positive for istradefylline, which indicates that istradefylline has a clinical effect unlikely to be attributed to chance. Study-level data for the change from baseline in ON time without troublesome dyskinesia to Week 12 for each of the dose levels of istradefylline compared to placebo is presented in Figure 45. These results further support the efficacy of istradefylline.

Figure 45: LS Mean Difference from Placebo in the Change from Baseline to Week 12 hours/day in the ON State without Troublesome Dyskinesia [95%CI] for Istradefylline (20 mg/day and 40 mg/day) - (OC, ITT, MMRM)



Source: Module 5, Re-exam Figure 9.2.

In the MAA submission, several subgroup analyses were conducted at the study level based on baseline factors such as age, race, gender, and the concomitant use of other drugs for PD. These analyses support internal consistency and the robustness of the findings for the primary endpoint and key secondary endpoint.

Eleven of the 12 istradefylline versus placebo comparisons for UPDRS III in the ON state are numerically positive for istradefylline. It is important to note that data on OFF time and ON time without troublesome dyskinesia are taken from patient diaries, while the UPDRS III outcome is based on the clinical assessment of motor signs of PD by the physician; UPDRS III data provide evidence that clearly supports the presence of a beneficial drug effect independent of the diary-based, patient reported primary and key secondary endpoints. Finally, the corresponding study-level data on UPDRS III are also presentedas part of the grounds of re-examination (data not shown in the CHMP AR)..

Returning to the pre-specified study level primary endpoint based on OFF time, the CHMP position is that 4 of the 8 trials met their primary endpoint although the sponsor argues that 5 out of 8 trials met their primary endpoint when Study 6002-US-006 is included, as justified under Ground 3. Looking across the trial program, there are no identifiable reasons why some trials did not meet the primary endpoint (if the drug is effective), nor are there obvious reasons why other trials did if the drug is ineffective (such as breaking of the blind, study conduct etc.); the replication of the positive results of istradefylline 20 mg/day, as well as the positive findings at 40 and 60 mg/day (one of the doses used in Study 6002-US-006) according to the pre-specified primary endpoint strongly indicate that the drug is effective in treating motor fluctuations in PD. The probability of 4 or more out of 8 trials giving a statistically significant result by chance alone is 0.000025[2] if the drug was indeed an inert substance. The probability of seeing 5 or more studies out of a program of 8 studies with a statistically significant result by chance

[1] Study 6002 US 018 also randomized subjects to istradefylline 10 mg/day.
[2] Binomial probabilities with a total of 8 trials and event (statistical significance) probability of 0.025.

with an inert compound is 0.00000051[1]. These probabilities establish strong support for the presence of a treatment difference for istradefylline compared to placebo. It is inconceivable that these results would be seen with an inert drug. The applicant has conducted a meta-analysis, which considers the totality of evidence from the pivotal study program for the treatment differences for reduction in OFF time and may therefore be more appropriate than counting and balancing formally positive vs formally negative studies.

Additional Support for Certainty of Effect

Random Effects Meta-analysis

Random effects meta-analysis is the accepted approach for combining results from a series of studies to gain an overall view of the body of evidence supporting the efficacy of a particular treatment and to obtain a more precise estimate of the treatment effect than any of the individual studies can provide. The applicant undertook a retrospective meta-analysis of the 8 pivotal studies to provide further evidence of the efficacy of istradefylline. Details are provided further below on the justification for the use of this methodology in this context.

Table 135 shows results for both random and fixed effect meta-analyses across the program of 8 studies for each of the 3 key endpoints under consideration in this Ground.

Table 135: Meta-Analysis Results: Difference from Placebo in the Change from Baseline to Week 12 for Hours in OFF time, Hours in ON time without Troublesome Dyskinesia and UPDRS III for 20 mg/day and 40 mg/day (OC, ITT, MMRM, Pool E1)

		Istradefylline 20 mg/day			**Istradefylline 40 mg/day**		
		Difference in LS means	**95% CI**	**p-value**	**Difference in LS means**	**95% CI**	**p-value**
OFF time (hours)	Fixed	-0.44	-0.67, -0.21	0.0002	-0.43	-0.66, -0.20	0.0002
	Random	-0.45	-0.75, -0.15	0.0031	-0.46	-0.80, -0.12	0.0074
ON time without troublesome dyskinesia (hours)	Fixed	0.43	0.16, 0.70	0.0015	0.31	0.06, 0.57	0.0158
	Random	0.43	0.14, 0.73	0.0039	0.34	-0.01, 0.69	0.0536
UPDRS III	Fixed	-0.87	-1.53, -0.21	0.0101	-1.28	-1.93, -0.64	0.0001
	Random	-0.87	-1.53, -0.21	0.0101	-1.27	-2.07, -0.47	0.0019

CI = confidence interval; UPDRS III = Unified Parkinson's Disease Rating Scale part III.
Source: Module 5, Day 120 Table 1.13.1, Table 1.13.2, and Table 1.13.3.

The random effects meta-analyses for LS mean change from baseline in hours OFF time indicates a beneficial effect for istradefylline at 20 mg/day and 40 mg/day when compared to placebo. The corresponding results for hours ON time without troublesome dyskinesia and the UPDRS III results are also supportive of the beneficial effect of istradefylline at 20 mg/day and 40 mg/day when compared to placebo.

These analyses provide additional evidence confirming the efficacy of istradefylline at both dose levels considered and for each of these key endpoints. In most cases, the results were statistically significant with CIs that were well separated from zero, clearly indicating a treatment difference.

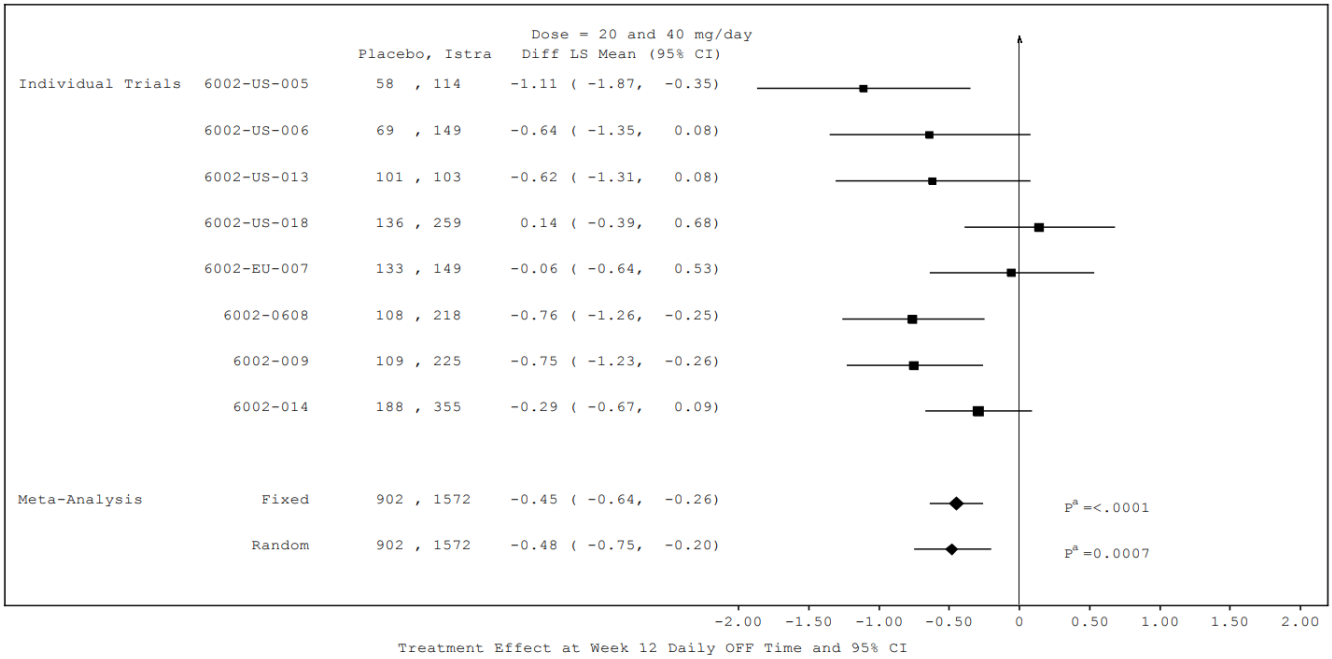
The applicant asserts that the data at the individual trial level and the meta-analysis, taken together, are sufficient to establish the certainty of a positive treatment effect of istradefylline across the pivotal program.

Combined Istradefylline 20 mg/day and 40 mg/day doses

It is accepted that the istradefylline 20 mg/day and 40 mg/day dose levels offer similar benefits in terms of the LS mean change from baseline in OFF time and corresponding LS mean change from baseline in ON time without troublesome dyskinesia differences from placebo, and data for these dose levels have been combined. This allows a more robust evaluation of the certainty of effect and provides a more precise estimate of the magnitude of that effect. Combining the 2 doses provides a test of whether active substances are efficacious and is an appropriate approach to demonstrate that istradefylline is efficacious. The test is not to replace evidence at individual dose levels, but is done in addition to describe the totality of evidence in favor of an effect of the active substance.

Figure 46 presents the results for each of the 8 studies in the istradefylline pivotal study program for difference in LS mean change from baseline to Week 12 in OFF time for istradefylline 20 mg/day and 40 mg/day combined compared to placebo together with the fixed and random effect meta-analyses.

Figure 46: LS Mean Difference from Placebo in the Change from Baseline to Week 12 hours/day in the OFF State [95%CI] for Istradefylline (20 and 40 mg/day Combined) - (OC, ITT, MMRM, Pool E1)



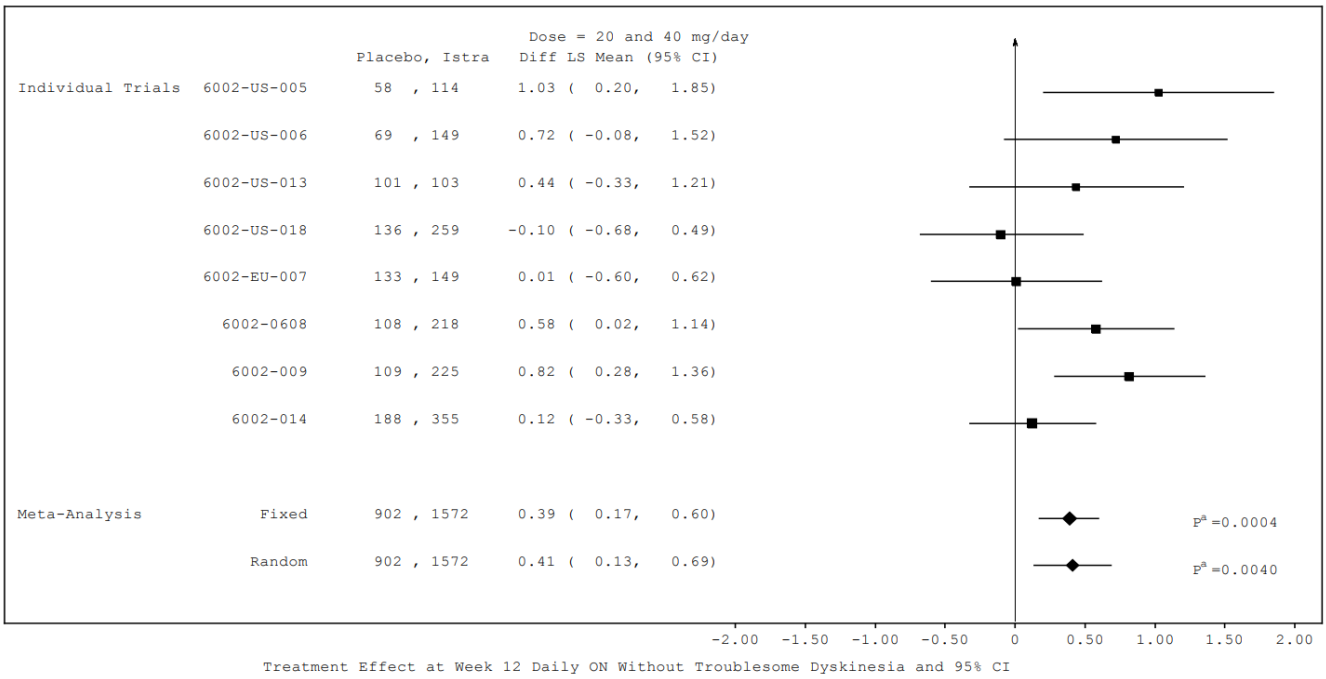
Source: Module 5, Re-exam Figure 2.1.1.
Note: 20 mg/day and 40 mg/day doses of istradefylline combined for Studies 6002-US-018, 6002-009, 6002-0608, and 6002-014.

The patterns seen in the forest plots at the individual study level for the istradefylline 20 mg/day and 40 mg/day dose levels combined are similar to the patterns seen at the individual dose levels, but with narrower CIs in those studies that included both dose groups. The random effects meta-analysis for the change from baseline in OFF time (istradefylline 20 mg/day and 40 mg/day combined) gives a point estimate of -0.48 hours (95% CI: -0.75 hours to -0.20 hours; p=0.0007). This result provides highly

statistically significant evidence for a treatment benefit for istradefylline (20 mg/day and 40 mg/day combined) compared to placebo. The I^2 statistic for heterogeneity was 48.24% (p=0.0603) (Module 5, Re-exam Table 2.1.1).

Figure 47 presents results for the key secondary endpoint, ON time without troublesome dyskinesia for istradefylline 20 mg/day and 40 mg/day dose levels combined compared to placebo.

Figure 47: LS Mean Difference from Placebo in the Change from Baseline to Week 12 hours/day in the ON State without Troublesome Dyskinesia [95%CI] for Istradefylline (20 and 40 mg/day Combined) - (OC, ITT, MMRM, Pool E1)

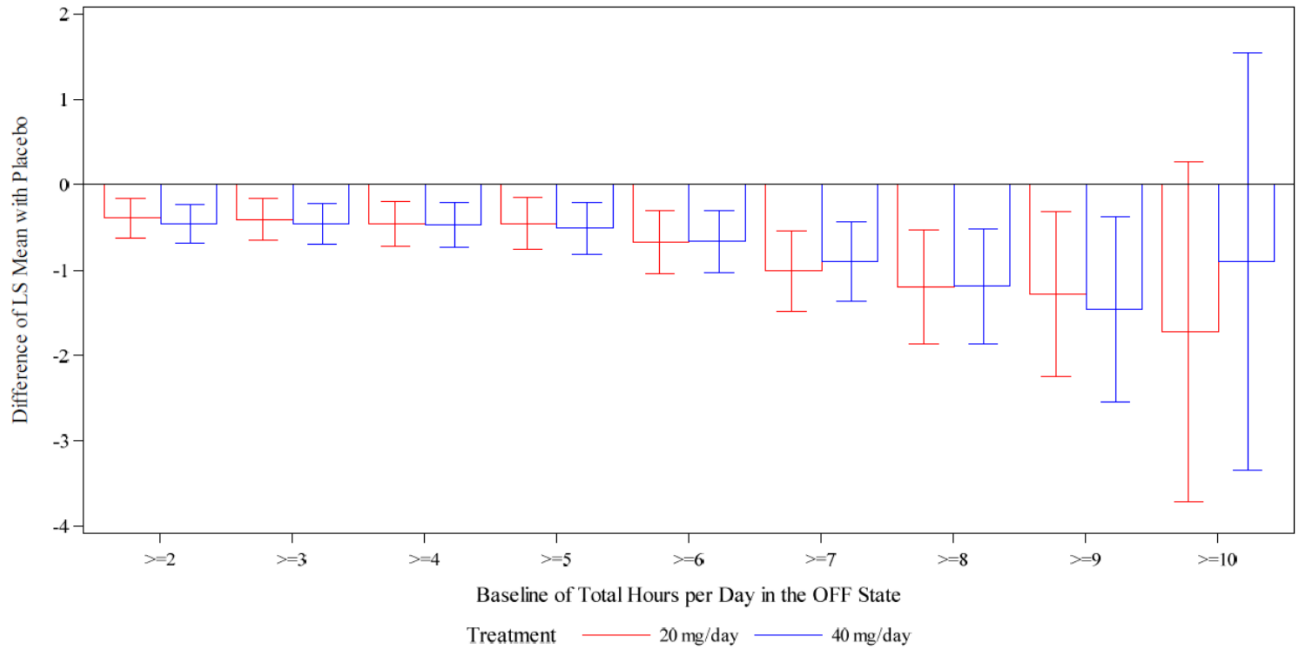


Source: Module 5, Re-exam Figure 2.2.1.
Note: 20 mg/day and 40 mg/day doses of istradefylline combined for Studies 6002-US-018, 6002-009, 6002-0608, and 6002-014.

1. Again, generally narrower CIs are seen for the combined dose groups at the individual study level. The random effects meta-analysis for the change from baseline in ON time without troublesome dyskinesia gives a point estimate of 0.41 hours (95% CI: 0.13 hours to 0.69 hours; p=0.0040), clear evidence supporting a benefit for istradefylline (20 mg/day and 40 mg/day combined) against placebo for this endpoint. The I^2 statistic for heterogeneity was 38.38% (p=0.1237) (Module 5, Re-exam Table 2.2.1).
2. The meta-analyses for each of the 20 mg/day and 40 mg/day dose levels of istradefylline and for the doses combined demonstrate a statistically significant benefit of istradefylline over placebo for both OFF time reduction and ON time without troublesome dyskinesia increase with p-values much smaller than the usual 0.05 cut-off for statistical significance and CIs that are clearly well away from the no treatment effect point.
3. These results confirm the certainty of a positive treatment effect observed across the individual studies for these 2 key endpoints.
4. Analysis by Baseline OFF Time
5. Several additional analyses were undertaken to further explore the certainty of effect of istradefylline. These analyses are post-hoc but are provided to further support a robust conclusion of efficacy.

In extensive subgroup evaluations presented in the Day 120 Subgroup Analysis Report, baseline OFF time was identified as the main effect modifier (treatment × covariate interactions: istradefylline 20 mg/day: p=0.0023; istradefylline 40 mg/day: p=0.0165). Figure 48 presents the treatment differences for each of the istradefylline 20 and 40 mg/day doses compared to placebo, cumulatively, according to baseline OFF time thresholds from 2 hours upwards.

Figure 48: LS Mean Difference from Placebo in the Change from Baseline to Week 12 Hours/day in the OFF State [95%CI] for Istradefylline (20 mg/day and 40 mg/day) According to Baseline Hours of OFF Time - (OC, ITT, MMRM, Pool E1)



Source: Module 5, Re-exam Figure 5.1.

Table 136 provides results of the subgroups associated with Figure 48. Complete results for the complementary subgroups associated with this table are provided as part of the grounds of re-examination (data not shown in the CHMP AR).

Table 136: Difference from Placebo in LS Mean Change from Baseline to Week 12 Hours/day in the OFF State [95%CI] for Istradefylline (20 mg/day and 40 mg/day) According to Baseline Hours of OFF Time - (OC, ITT, MMRM, Pool E1)

	Placebo		Istradefylline 20mg/day			Istradefylline 40mg/day		
	N	LS mean change from baseline (95% CI)	N	LS mean change from baseline (95% CI)	Difference from placebo in LS mean change (95% CI)	N	LS mean change from baseline (95% CI)	Difference from placebo in LS mean change (95% CI)
Baseline Subgroup ≥ hours/day in the OFF state								
≥2	990	-0.83 (-0.98,-0.67)	845	-1.22 (-1.39,-1.04)	-0.39 (-0.62,-0.16)	875	-1.28 (-1.45,-1.11)	-0.46 (-0.68,-0.23)
≥3	940	-0.91 (-1.08,-0.75)	790	-1.32 (-1.51,-1.14)	-0.41 (-0.65,-0.16)	810	-1.38 (-1.56,-1.20)	-0.46 (-0.70,-0.22)
≥4	832	-1.01	691	-1.47	-0.46	720	-1.48	-0.47

		(-1.19,-0.83)		(-1.67,-1.27)	(-0.72,-0.19)		(-1.68,-1.28)	(-0.73,-0.21)
≥5	684	-1.13 (-1.34,-0.92)	567	-1.59 (-1.82,-1.35)	-0.46 (-0.76,-0.15)	584	-1.64 (-1.87,-1.42)	-0.51 (-0.82,-0.21)
≥6	522	-1.33 (-1.58,-1.08)	420	-2.00 (-2.29,-1.72)	-0.67 (-1.04,-0.30)	444	-1.99 (-2.27,-1.71)	-0.66 (-1.03,-0.30)
≥7	365	-1.42 (-1.74,-1.11)	279	-2.43 (-2.80,-2.07)	-1.01 (-1.48,-0.54)	314	-2.32 (-2.67,-1.97)	-0.90 (-1.36,-0.43)
≥8	233	-1.73 (-2.17,-1.28)	182	-2.93 (-3.44,-2.41)	-1.20 (-1.87,-0.53)	190	-2.91 (-3.41,-2.42)	-1.19 (-1.86,-0.52)
≥9	132	-2.02 (-2.68,-1.37)	127	-3.30 (-4.00,-2.61)	-1.28 (-2.25,-0.31)	106	-3.48 (-4.27,-2.69)	-1.46 (-2.55,-0.37)
≥10	71	-2.46 (-3.76,-1.17)	64	-4.18 (-5.47,-2.89)	-1.72 (-3.71,0.27)	52	-3.36 (-5.01,-1.71)	-0.90 (-3.34,1.54)

CI = confidence interval; LS = least squares. Pool E1 includes Studies 6002-US-005, 6002-US-006, 6002-US-013, 6002-0608, 6002-009, 6002-US-018, 6002-EU-007, and 6002-014.
Source: Module 5, Re-exam Tables 4.1.1 to 4.9.1.

The LS mean change from baseline in hours of OFF time to Week 12 increases in magnitude as baseline OFF time increases in each istradefylline treatment group, meaning that those with the greatest unmet need in terms of OFF time at baseline are deriving the most benefit. Although this pattern is seen in the placebo subgroups, the magnitude of increasing response with increasing baseline OFF is larger in the istradefylline cohorts. This relationship between treatment benefit and baseline OFF time clearly supports the presence of a genuine drug effect over and above any regression to the mean. Were istradefylline to have no true effect, a pattern of outcomes as displayed would be extremely unlikely to occur. This relationship between treatment benefit and baseline OFF time clearly supports the presence of a genuine drug effect. Were istradefylline to have no true effect, a pattern of outcomes as displayed would be extremely unlikely to occur.

Certainty of the Magnitude of Effect

The random effects meta-analysis for the combined istradefylline 20 mg/day and 40 mg/day doses gives a treatment effect of -0.48 hours (95% CI: -0.75 to -0.20) in the change from baseline for hours OFF time compared to placebo. The upper limit of this CI is approaching 50% of the point estimate in terms of the distance from zero and is clearly not in close proximity to the no treatment effect point. With 97.5% confidence this meta-analysis shows that the treatment effect size for the absolute reduction if OFF time is at least 0.20 hours is in favor of istradefylline.

It is not the case that every value in the CI is supported equally by the data. Values at the lower end (and at the upper end) have less support than values towards the middle of the CI. The argument that the true effect could well be below the point estimates of -0.48 is countered by equal likelihood that the true effect could be larger.

Interpretation regarding the magnitude of a treatment effect should be based on the point estimate of that effect. The point estimate (-0.48 hours, in this case) is the value for the treatment difference that is recognized as the best estimate of the treatment effect based on the data.

Consistency of Effect

The CHMP position is that 4 of the 8 trials met their primary endpoint while 4 trials failed to do so, and it is this aspect (4 successful studies and 4 failed studies) that has possibly led to the impression that the results from the istradefylline trial program are inconsistent. In this section, consistency is considered more appropriately, by looking at the magnitude of treatment effects across the program. Initially, these calculations will be undertaken for the combined istradefylline 20 and 40 mg/day groups with subsequent comments on the corresponding results for the separate dose levels.

The observed treatment effects for the change from baseline in OFF time are provided in

Figure 46 for the combined 20 mg/day and 40 mg/day dose groups compared to placebo. The corresponding treatment effects for the change from baseline in ON time without troublesome dyskinesia, are provided in Figure 47.

Summary data for the sample sizes and inter-patient variability, by treatment group and by study (Pool E1), for the change from baseline in OFF time to Week 12 are also provided as part of grounds for re-examination (data not shown in the CHMP AR).. Patient-to-patient variability is measured by the SD. Similar data for ON time without troublesome dyskinesia are provided as part of the grounds of re-examination (data not shown in the CHMP AR)..

The average sample size for placebo was 122, while the average sample size for istradefylline 20 mg/day and 40 mg/day groups combined was 211. For both endpoints, the inter-patient variability is quite similar across treatment groups and across studies. Averaging the SDs across the pivotal study program and placebo, 20 mg/day and 40 mg/day istradefylline treatment groups gives a SD of 2.51 hours for the change from baseline in OFF time and a SD of 2.73 hours for the change from baseline in ON time without troublesome dyskinesia.

Note that this SD for the change from baseline in OFF time is entirely in line with those in Figure 1 of the Stowe et al (2010) publication. This level of inter-patient variability is an inherent aspect of PD and data collected through diary cards and is a key aspect of the recognized heterogeneity within this disease area.

In a series of identical trials where the true treatment effect in change from baseline in OFF time is equal to -0.48, 95% of the observed study treatment effects will lie between -1.05 hours to +0.09 hours. Note that the value -0.48 hours chosen here is the value resulting from the random effects meta-analysis, which provides the best estimate of the true effect size for istradefylline 20 and 40 mg/day doses combined. This calculation is based on assuming that treatment effects have a normal sampling distribution with mean -0.48 hours and SD given by the observed treatment effect standard error (SE; $2.51\sqrt{(1/122+1/211)}$). The 95% CI is then calculated as -0.48 ± 2*SE.

The treatment effects observed in the trials are within this range with 2 exceptions. Study 6002-US-005 has an observed treatment effect of -1.11 hours, while Study 6002-US-018 has an observed treatment effect of 0.14 hours, values which are just outside of the predicted range in both directions. The 8 trials in the program are not identical, with slightly different protocols with, for example, minor differences in the inclusion/exclusion criteria, different numbers of treatment groups, small differences in trial duration, and so on; a certain amount of extra variability will result from these differences. Based on these considerations, a level of consistency is seen across the program, which is exactly in line with what one would expect to see in a series of studies of this kind, based on a true treatment effect of -0.48 hours and inherent inter-patient variability as seen within the trials on average. This analysis was repeated for the individual dose levels and the overall conclusions remained the same[3]. Most of the individual study treatment effects were within the ranges predicted based on the observed treatment effect from the

[3] Intervals: 20 mg/day, -1.07 to +0.17 based on treatment effect size of -0.45 hours, 40 mg/day, -1.08 to +0.16 based on treatment effect size of -0.46 hours.

meta-analysis for that dose, an assumed common value for the SD for that endpoint, and sample sizes averaged across the placebo, istradefylline 20 mg/day and istradefylline 40 mg/day dose groups.

Assuming a series of identical trials with the same sample sizes (and here we are using an average sample size of 122 for the placebo group and an average sample size of 141 for istradefylline at 20 mg/day or 40 mg/day) and inter-patient variability as measured by a SD of 2.51, a treatment effect of approximately -0.62 hours would be needed for statistical significance to be achieved for an istradefylline 20 mg/day or an istradefylline 40 mg/day comparison with placebo. It is therefore not surprising that some studies failed to meet the requirement with observed treatment effects that did not reach the required -0.62 hours for statistical significance. But importantly, the studies in the program do show consistency around the overall estimated treatment effect.

Of note, the actual treatment effects seen in the program align with the -0.62 cut-off only approximately, given the different SDs and sample sizes used in the individual studies.

Similar considerations for ON time without troublesome dyskinesia support the consistency in the effect of istradefylline across the 8 studies for this key secondary endpoint. In a series of identical trials where the true effect (change from baseline in ON time without troublesome dyskinesia) is 0.41hours, 95% of the observed treatment effects for the combined istradefylline 20 mg/day + 40 mg/day dose groups will lie between -0.21 hours to +1.03 hours. For this endpoint, the treatment effect sizes are within this range in all 8 trials. Again, there is no support for a conclusion of inconsistency of outcome across the istradefylline trial program.

As demonstrated in this section, when the predicted range of effects across the istradefylline program is calculated based upon the most statistically certain estimate of the mean treatment effect of istradefylline, the apparent inconsistency in results is overcome. The variation in study results observed across the istradefylline pivotal study program are largely consistent with inherent random fluctuation and are reflective of the heterogeneity seen in PD, in PD programs (Stowe, 2010; Li, 2017) and in CNS disease programs more widely.

Justification of Retrospective Meta-Analysis

The CHMP have indicated that the pooling of data cannot be used to establish the efficacy of istradefylline. However, the CHMP Points to Consider on Application with 1. Meta-Analyses; 2. One Pivotal Study states that there are situations where the need for a meta-analysis becomes apparent only after the results from some, or sometimes all, studies are known, for example, when there is a need to put seemingly inconsistent results into perspective. This is the case with istradefylline, where the CHMP's view is that there are 4 positive and 4 negative studies. The meta-analyses consider the totality of evidence on the primary efficacy variable, change from baseline in OFF time to Week 12, and these results reinforce the conclusion that istradefylline is efficacious and provide a precise estimate of the magnitude of the benefit of istradefylline.

The CHMP Points to Consider paper outlines conditions under which such an analysis will be of value, stating that the prerequisites for a retrospective meta-analysis to provide sufficient evidence for a claim include:

1. Some studies clearly positive.
2. Inconclusive studies showing positive trends in the primary variable.
3. No statistically significant heterogeneity.
4. Pooled 95% CI well away from zero.
5. A justification that a biased selection of studies and/or endpoints is unlikely.
6. A sensitivity analysis demonstrating robustness of the findings.

These points are addressed in turn below:

1. Some studies clearly positive.

   Four studies are positive according to the pre-specified primary endpoint and method of analysis. Note however that the applicant views a fifth Study 6002-US-006 as also positive for istradefylline (see Ground 3).

2. Inconclusive studies showing positive trends in the primary variable.

   Of the 3 studies that did not reach statistical significance, 2 of 3 gave numerical positive outcomes for istradefylline. The single trial that had point estimates not in favor of istradefylline for the change from baseline in OFF time was Study 6002-US-018.

3. No statistically significant heterogeneity.

   The statistical test for heterogeneity gave p=0.1657 for istradefylline 20 mg/day and p=0.0626 for istradefylline 40 mg/day, both non-significant at the 5% level.

4. Pooled 95% CI well away from zero.

   The difference in LS means in change from baseline in the total hours per day in the OFF state was -0.48 hours (95% CI: -0.75 to -0.20; p=0.0007) for the combined istradefylline 20 and 40 mg/day doses versus placebo. The upper limit of this CI is approaching 50% of the point estimate in terms of the distance from zero and is clearly not in close proximity to the no treatment effect point.

5. A justification that a biased selection of studies and/or endpoints is unlikely.

   A biased selection of studies and/or endpoints is unlikely as Pool E1 included all 8 controlled Phase 2b/3 trials (Studies 6002-US-005, 6002-US-006, 6002-US-013, 6002-009, 6002-0608, 6002-US-018, 6002-EU-007, and 6002-014) and the primary endpoint used in the meta-analysis was consistently defined across the trial program. The applicant had access to all data from each of these studies and those data were used in all analyses. All studies were well-controlled and conducted in accordance with GCP.

6. A sensitivity analysis demonstrating robustness of the findings.

   For the pooled analyses, several different strategies were used to deal with missing data, for example, an observed case analysis treated all invalid timepoints for the diaries as missing, and a worst-case analysis imputed OFF for missing timepoints for the active treatment groups and imputed ON without dyskinesia time for the placebo group. In each case, the pattern of results remained largely unchanged.

The Points to Consider paper goes on to state that to minimize the opportunity for a retrospectively specified meta-analysis protocol to be data dependent, the primary specifications and definitions set up in the individual studies should be followed. The use of the random effects meta-analysis was not pre-specified. However, a closely related methodology using pooling of the data and associated modelling was discussed with the regulatory authorities at pre-submission meetings. This pooling/modelling strategy was detailed in the MAA SAP and aligns closely with the fixed effect meta-analysis reported here during the evaluation prior the CHMP Opinion on 22 July 2021. Random effects meta-analysis is recognized as a more conservative approach compared to fixed effect meta-analysis. Furthermore, it is clear that the results presented for the fixed effect meta-analyses are in line with those based on random effects meta-analyses.

Robustness of Meta-Analysis

The MAA Statistical Analysis Plan was provided at the time of the initial submission, and clearly stated that there would a 'pooled' analysis across all 8 studies (Pool E1). The primary endpoint in the pooled

analysis that was discussed in the pre-submission meetings was the change from baseline in hours OFF time. This was the primary endpoint in some of the studies. In other studies, the OFF-time endpoint was the change from baseline in percentage of awake time spent in the OFF state. Meta-analysis results for the change from baseline in percentage of awake time spent in the OFF state are presented as part of the grounds of re-examination (data not shown in the CHMP AR) and give results and conclusions that are entirely in line with the results for the change from baseline in hours OFF time.

The meta-analyses were compiled from study results that were themselves based on individual patient data. The applicant had access to all data, and this facilitated consistency in data handling and the specification and application of statistical modelling.

Statistical Modelling

A consistent approach was used in the modelling across all studies as detailed in MAA SAP and this is aligned with how the meta-analyses were conducted. The primary timepoint for each variable was Week 12 and data were analyzed using a MMRM approach, with baseline value as a covariate, fixed effect terms in the model for pooled study center, treatment group, week, and treatment-by-week interaction, and an unstructured covariance matrix. Rules were developed for combining sites, and no questions were raised by the assessors in relation to the validity of this model during pre-submission meetings. Using this model, the difference in change from baseline in total hours OFF time between each istradefylline dose group and placebo was estimated based on the LS mean difference with a corresponding 95% CI and p-value. The robustness of results based on the MMRM model were evaluated by using sensitivity analyses which relaxed the assumptions on which that model is based.

This random effects meta-analysis methodology was chosen as the basis for combining results across studies during the evaluation procedure prior to CHMP opinion on 22 July 2021. This method is the method of choice (Cochrane Handbook for Systematic Reviews of Interventions) to incorporate trial-to-trial heterogeneity. It is a conservative approach compared to fixed effect meta-analysis and modelling based on pooled data, and directly account for the potential for additional heterogeneity in treatment effects across the trials.

Strength of Evidence from the Meta-Analysis that Confirms Efficacy

The Points to Consider paper on meta-analyses state that in cases where the meta-analysis provides the pivotal evidence for an indication, a p-value more extreme than the conventional significance level of 0.05 would generally be required, and a narrower CI for the treatment effect would be expected compared to that from a single trial. The required degree of significance will be judged on a case-by-case basis considering factors such as the amount of supportive data, plausibility of the hypothesis tested, and whether the analysis is pre-specified or not. These analyses provide additional evidence that can also be considered as further confirming the efficacy of istradefylline at both 20 mg/day and 40 mg/day separately and combined, for the primary endpoint. The meta-analysis results show a high level of statistical significance with CIs that are well separated from zero, clearly indicating a treatment difference.

The SAP within the MAA dossier. The rationale for the pooling and the pooling methodology was discussed with the assessors in pre-submission meetings. The analysis of the combined doses of istradefylline across all 8 studies, as shown in

Figure 46 has a p-value for the change from baseline in OFF time compared to placebo of 0.0007, which is clearly more extreme than the conventional significance level of 0.05, and the CIs are narrow (-0.75, -0.20). The upper limit of this CI is clearly not in close proximity to the no treatment effect point. The amount of supporting data is strong (a low p-value is also seen for the analysis of ON time without troublesome dyskinesia [p=0.0040]) and the hypotheses tested are clearly plausible. The treatment

effects seen for the important secondary endpoint, UPDRS III, are also supported by the associated meta-analysis at both istradefylline 20 mg/day and 40 mg/day dose levels.

Overall, the applicant concludes that the meta-analysis conducted is valid, appropriate, consistent with CHMP Points to Consider paper, and substantiates the efficacy of istradefylline demonstrated in the pivotal study program.

CHMP discussion on ground for re-examination #1

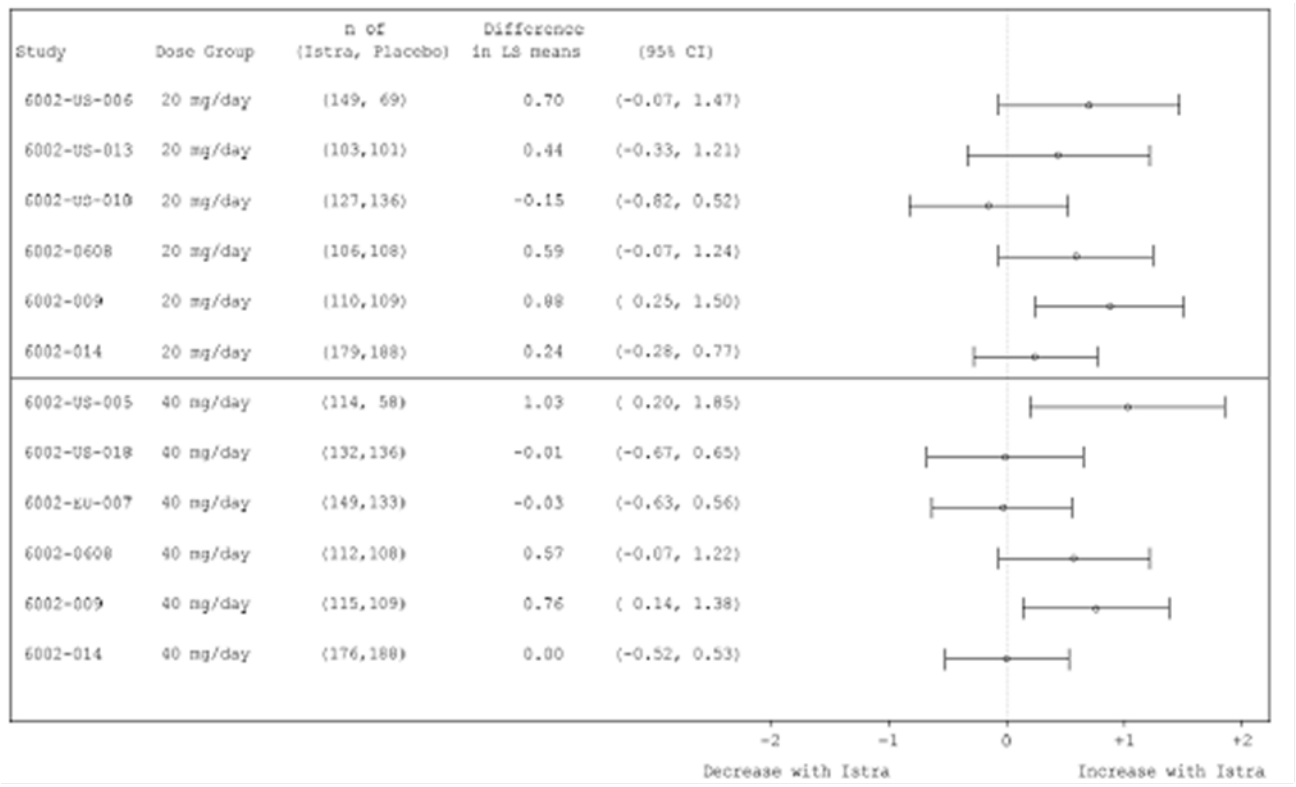
Background to the re-examination

The benefit/risk of istradefylline needs to be re-examined based on the totality of the data including the results of eight pivotal trials and the combined data from the post-hoc meta-analysis on the pooled patient data from the eight pivotal studies.

The effect of istradefylline on reducing the awake time per day spent in the OFF state is uncertain.

There was a positive signal in USA studies 005 and 013 (primarily expressed as percentage of awake time OFF) and in Japanese studies 0608 and 009 (primary endpoint: hours/day in the OFF state). USA studies 006 and 018 (percentage of awake time OFF) and European studies 007 (% awake time OFF) and 014 (hours/day in the OFF state) did however not provide confirmation of a treatment effect. Also the results for the two doses are divergent between studies so that neither a clear dose-response relationship could be established, nor could even any dose prove to be consistently efficacious (Figure 44). The beneficial effects of istradefylline are questioned also when judged from the results of the relevant secondary outcome measure hours/day in the ON state without troublesome dyskinesia (Figure 49).

Figure 49: Difference in LS Means (95% CI) (vs. placebo). Change from baseline at week 12 total hours per day ON without troublesome dyskinesia by treatment MMRM- Observed Case Analysis – ITT Analysis set- 8 studies.



The consistency across the program has been claimed by the applicant as similar to that expected in studies of this kind as well as the inherent inter-patient variability. This is not fully understood and it does not seem to be the case for the clinical development of medicinal products approved in Europe for a similar population with respect to clinical and demographic characteristics (e.g rasagiline [Azilect®], opicapone [Ogentys®], safinamide [Xadago®]. The evidence is in general substantiated by the replicated clinically and statistically significant effect of two positive trials in which the dose-response relationship is more clearly suggested. Whereas the clinical developments of these products are not identical and with all the intrinsic limitations of indirect comparisons the inconsistencies observed in the results for istradefylline, do not appear to have occurred to the same extent in the named approved treatments.

Re-examination assessment

The original application contains eight pivotal clinical studies, with four positive studies and four negative studies. The applicant argues that the chance that four out of eight studies are positive by chance alone is 0.000025. Rather than the view on the trial, the more relevant view is on the individual doses on which 3 out of 6 comparisons were successful. Applying the same combinatorial calculations a probability of around 0.0003 (assessor calculation) results for each dose. However, if there is a small effect, lower than that anticipated in the sample size calculation, it would not be unlikely to find four out of eight studies being statistically significant. The question then is, whether the effect is clinically relevant. Nevertheless, the fact remains that there are also four negative studies. Moreover, in this calculation all studies are treated the same (study size, effect estimates, distance to threshold etc. are ignored) and applying this dichotomy is not a meaningful way to assess efficacy. From the methodological perspective the 3 out of 6 comparisons (or 4 out of 8 studies) criterion is also not the predefined way of concluding on efficacy. Therefore, while the applicants point is acknowledged, the discussion of the pooled analysis is of more informative value (see below), and the same is also concluded by the applicant.

The applicant also argues that the treatment effect of most studies (6 out of 8) fall within the range of expected treatment effects. However, this range is calculated on the same eight studies and should not be a surprise. This is considered circular reasoning and is not considered a proper argument for consistency of effect. Nevertheless, even two of the 8 comparisons fail to be within the prospected range. Referring to Figure 46 (which the applicant uses for their justification), it can be seen that the CIs for the two most extreme studies US-005 and US-018 do hardly overlap. Moreover, the inconsistency of effect does not allow to conclude on a positive benefit/risk.

Given the inconsistency of the individual study results the applicant undertook a pooled analysis of the 8 pivotal studies. The applicant argues that their meta-analysis of the eight studies is sufficient based on the prerequisites in the CHMP Points to Consider on application with 1. meta-analysis; 2. one pivotal study (CPMP/EWP/2330/99):

1. Some studies clearly positive.
2. Inconclusive studies showing positive trends in the primary variable.
3. No statistically significant heterogeneity.
4. Pooled 95% CI well away from zero.
5. A justification that a biased selection of studies and/or endpoints is unlikely.
6. A sensitivity analysis demonstrating robustness of the findings.

However, this can be debatable on several of the prerequisites. The CPMP/EWP/2330/99 guideline states that "*A retrospective meta-analysis of only two studies originally intended to stand on their own is not expected to add any useful information. In particular, a meta-analysis cannot be used to reconcile the conflicting results of one positive and one inconclusive study.*" This remark can be extended to four positive and four inconclusive studies. However, since it appears to be to some extent at odds with the first two prerequisites, there is some room opened to consider a meta-analysis as an exceptional case, though such will always be problematic. The points to consider in addition also say that the necessity to resort to a meta-analysis might indicate that there is a reason to question clinical relevance, and a valid meta-analysis does not necessarily mean that an application is approvable. In that sense a prerequisite is a necessary, but not a sufficient condition, if the data are not convincing.

While it is agreed that some studies are at least positive based on statistical significance, the clinical relevance of these findings is debatable. While most inconclusive studies show positive trends, for the 20mg dose 1 trial estimates a negative effect, while for the 40mg dose 2 trials show no trend for an effect (i.e. the point estimate lies on the null hypothesis). It is not agreed that these are positive trends, and this requirement is not considered met.

Heterogeneity was tested by the applicant by I^2-test and corresponding p-values. According to the applicant heterogeneity was not statistically significant (p=0.1657 for 20 mg/day and p=0.0626 for 40 mg/day). However, the I^2 values (around 40-50%) did indicate moderate heterogeneity. Generally, such tests start with the null-hypothesis of no heterogeneity, and are usually not very powerful to detect differences. As a consequence, a non-significant result must not immediately be taken as evidence of no heterogeneity. Furthermore, it is not uncommon to use a threshold of at least 0.10 to detect heterogeneity, rather than the conventional level of 0.05. Although additional calculations are provided to advocate the likelihood of a common mean and SD in all studies the proper assessment of heterogeneity is the forest plot and I^2 value. Nevertheless, the applicant did perform a random effects model in the response, which should account for (part of) the heterogeneity and could be considered as the standard approach.

Whether the lower bounds for the two pooled 95% CI are or are not far from the null has a methodological

and a clinical component. The argumentation on this prerequisite is not agreed to and is not considered fulfilled. In the random effects meta-analysis model lower limits of the confidence interval of -0.15hrs (9min) for the 20mg dose and -0.12 (7.2min) for the 40mg dose are estimated. This is not considered to be well away from zero. In addition to this, the discussion below that these are nominal levels that cannot be interpreted as what has been demonstrated must be noted.

The company argues that these lower limits are about halfway the null and the point estimate and thus "well away". Such an argument does not imply that effects are clinically relevant as for any non-zero mean and standard deviation a sample size can be found where the lower limit of the 95% CI is expected to lie halfway between the null and the mean. In this respect it must also be noted that the pooled analysis is based on a total of eight studies which where all powered individually to detect clinically relevant effects, resulting in narrowing the width of the 95% CI as compared to individual trials and increasing the power for detection of small (potentially clinically non-relevant) effects. Pooling of 8 equally-sized studies would enable detection of effect sizes that are almost three times smaller than those detectable in the separate studies. Notably, the upper limit is well away from the established minimal clinically important difference (MCID) of 1 hour. Moreover, while the p-values are low when compared to the standard 0.05, if the meta-analysis is used as the pivotal evidence (and thus a single pivotal study) a stricter threshold is usually required.

It is agreed that there is no biased selection of studies, as long as all eight studies are included, and that the meta-analysis results do appear robust.

In summary, the applicants conclusion that all prerequisites for the acceptance of the retrospective meta-analysis is disagreed. Especially, the point that the results are well away from zero is not agreed to. The nominal 95% confidence intervals would represent an improvement in off time of only 9min resp. 7.2 min for the two doses, and this is considered minimal, but not well away from 0.

The SAP is dated 3 September 2019, well after finalisation of the studies. Furthermore, this SAP could not follow the analysis plans of the individual studies: The primary endpoint was not identical across studies and also the number of diary days used for the endpoints differed. Initially a fixed effects model and later - given the heterogeneity of the studies due to their regional and wide temporal spread - a more appropriate random effects model was used. What is hampering the interpretation of the meta-analysis, is that it was performed retrospectively, in order to rescue the initial pathway that was based on the assessment of the individual trials, but which failed to be convincingly establish efficacy. In addition, another important aspect is that the meta-analysis is essentially a one-pivotal trial, for which the standard alpha-level of 5%, two-sided (resp. the 95%-confidence interval) does not apply to demonstrate statistical significance, as is laid out in the same points to consider. These points to consider do not specify a specific level. It is considered that especially in the case of a post-hoc meta-analysis intended to rescue borderline results it is relevant not to fall below the level of evidence that is required in the standard case. The usual two-pivotal trials at a two-sided 5%-level paradigm translates in the one-pivotal trial setting equivalently to a one-sided alpha-level of 0.000625 (resp. a two-sided alpha-level of 0.00125) that will (maximally) need to be achieved. The random-effects model achieves a two-sided p-value of 0.0031 for the 20mg dose and a p-value of 0.0074 for the 40mg dose. The individual doses therefore both miss the equivalent that is usually applied in the standard two-pivotal trial paradigm.

Therefore, these post-hoc defined meta-analyses (which by definition further lack an applicable threshold and the p-values and confidence intervals are to be seen as nominal levels) miss the stringency that is applied in the standard setting. In addition, several elements contribute to that the achieved p-values resp. the levels to be achieved, can further be questioned. One is that two doses are tested in the two meta-analyses, but other than in the individual trials no multiplicity adjustment was (and could be) done. In light of no clear pattern of dose response the (hypothetical) choice for multiplicity control was not

obvious, and depending on the choice this would have further decreased the necessary p-value. Of further note is also that the pooled analysis of the data (other than a combination of the study results) takes advantage of the fact that the analysis model contains the baseline variable, which generates a more favourable result for the failed trial Study US-006.

The applicant also presents meta-analysis results for the pooled dose arms. This pooling was not predefined in the analysis plans of the separate studies and only as exploratory in the pooled analysis plans. Anyway, the results for the pooled doses is not considered relevant for the assessment of the individual doses and the argumentation that "it is accepted that the istradefylline 20 mg/day and 40 mg/day dose levels offer similar benefits" is in contradiction with the posology as presented in the SmPC, where both doses are proposed separately. Therefore, focus will be on the results of the separate dose arms.

Though considered problematic, there is some room for allowing a meta-analysis in exceptionally convincing cases and based on the prerequisites as described in the Points to consider. However, in the current application multiple aspects stand against following a conclusion following this route, e.g. lack of pre-definition of this post-hoc route, heterogeneity between studies, statistical rigour in terms of evidence level achieved, no multiplicity adjustment for the two doses, and (nominal) confidence limits that are not considered to be well away from zero.

Even if we would step over these problems of the meta-analysis, we are still left with the poor clinical relevance of the results. The overall effect size is considered meagre, and the lower limit of the 95% CI is considered close to the null. The applicant argues that it is only half way between the point estimate and the null, however, the lower bound is below the conventional MCID of 1 hour and is also below the MCID of 20 to 30 minutes the applicant concluded on based on patient surveys and expert opinion. Furthermore, the upper limit of the 95% CI does not even cross the conventional MCID of 1 hour. Taken into account that a larger sample size (as a consequence of meta-analytic pooling of different studies) would allow narrower confidence intervals, the current results are also not considered convincing. It is concluded that the standard pre-requisite, defined in EMA guidelines, of pooled 95% confidence interval well away from zero is not met.

The exploratory subgroup analysis according to baseline OFF time appears to show a larger treatment effect in patients with higher baseline OFF time. However, interpretation is hampered by large CIs. Therefore, the applicants argument that this indicates "a genuine drug effect" is not agreed. Another conclusion can be that patients with higher baseline values represent a population with suboptimal therapeutic control of OFF time at study entry (see comments on the additional ground for re-examination).

Concluding, the pooled analysis had not been predefined, but was rather performed as a rescue measure following the observation that the individual trials, which were planned to stand on their own, showed only borderline or no effect. Next to various methodological concerns, as mentioned above, the estimated effect size with the random effects meta-analysis was modest and while the 95%-CIs excluded '0', given the proximity of the CI-boundaries to 'no effect', a clinically relevant treatment effect cannot be confirmed. This precludes the use of the meta-analysis as pivotal evidence for the MAA in a situation where also the results from eight individual studies did not allow to conclude that efficacy was established due to inconsistent effects.

Point not resolved

5.1.2. Ground #2 (dot 3 of CHMPs GfR)

Grounds for re-examination:

The CHMP has not fully considered the evidence of effectiveness in Study 6002-014. The applicant contends that there is evidence of a positive treatment effect of istradefylline in 'maximally treated' patients and in patients from Europe.

Applicant's Position on Ground 2

- *Positive treatment effects in the pre-defined endpoints are evident in Study 6002-014 which should not be underestimated, despite the primary endpoint not meeting statistical significance.*
- *The OFF-time treatment effect for istradefylline in Study 6002-014 falls within the predicted range, given a number of factors including the interpatient variability, the estimated OFF time treatment effect and the sample size per treatment arm.*
- *Post-hoc analyses taking into account the magnitude and frequency of benefit over time provide further evidence of a treatment effect for istradefylline compared to placebo in this study.*
- *The applicant demonstrates that the timing of the study, the characteristics of maximal treatment and the inclusion of European patients in Study 6002-014 did not influence study outcome:*
    - *Standard of care has not changed throughout the pivotal study program. No new classes of drugs became available during this period.*
    - *A consistent positive istradefylline effect was observed across the regions.*
    - *The result in the Study 6002-014-like subgroup from Pool E1 shows nominal statistical significance.*

Discussion of Study 6002-014

Study 6002-014 was designed at the request of the US FDA, to conduct a trial in PD patients who were considered to be 'maximally and optimally treated' (i.e., a population with a higher dopaminergic load than the earlier istradefylline studies). The definition of 'maximally and optimally treated' was reflected in Study 6002-014 by 3 key inclusion criteria (in addition to end-of-dose wearing-off): documented levodopa-induced dyskinesia, use of levodopa at a dose of ≥400 mg/day and treatment with at least 1 additional dopaminergic PD medication (dopamine agonist, COMT inhibitors, MAO- B inhibitors). The levodopa threshold selected indicates a need to have increased the dose to maintain response duration as the patient's capacity to store dopamine diminishes as the disease progresses (Sveinbjornsdottir, 2016); the presence of dyskinesia reflects the development of motor complications as a result of the pharmacological dopaminergic load (Olanow et al, 2013); and administration of additional PD medications reflects the need to manage symptom fluctuations.

Study 6002-014 was conducted between November 2013 and September 2016 and enrolled patients to 88 global sites in the USA, Canada and Israel and in the following countries in Europe: Czech Republic, Germany, Poland, Serbia and Italy. The study randomized 613 subjects to receive istradefylline 40 mg/day (207 subjects), istradefylline 20 mg/day (202 subjects) or placebo (204 subjects).

The primary endpoint was the change from baseline in the total hours of awake time per day spent in the OFF state at Week 12 based on the 24-hour ON/OFF patient diary data. The differences in the LS mean changes from baseline between the istradefylline 20 mg/day and 40 mg/day groups and placebo were -0.32 hours and -0.27 hours, respectively. The pairwise treatment comparisons for each

istradefylline dose versus placebo based on MMRM were not statistically significant; p=0.156 (20 mg/day versus placebo) and p=0.234 (40 mg/day versus placebo), however the trends were clearly positive. Numerically positive, though non-significant results were also seen for the secondary endpoints, ON time without troublesome dyskinesia - 20 mg/day versus placebo (0.24 hours; p=0.366) and UPDRS Part III – 20 mg/day (-0.1; p=0.884) and 40 mg/day (-1.0; p=0.141) groups vs placebo, respectively. The exception was the ON time without troublesome dyskinesia treatment effect in the 40mg/day versus placebo analysis (0.0 hours; p=0.986).

With regard to the primary endpoint, it has been outlined within the applicant's position on Ground 1, that the treatment effect observed in Study 6002-014 for the combined dose groups (20 mg/day + 40 mg/day: -0.29 hours is within the predicted range (-1.05 to +0.09 hours), given the patient to patient variability (measured by the SD), the average sample size of the treatment arms across the pivotal study program and the magnitude of the estimated treatment effect of -0.48 hours for istradefylline (random effects meta-analysis for 20 mg/day+40 mg/day). Given these factors, it is not particularly surprising that the observed change from baseline in OFF time treatment effects in some studies, for example Study 6002-014 (observed treatment effect -0.29 hours), did not reach statistical significance. The corresponding intervals for 20 mg/day and 40 mg/day doses separately, given treatment effect sizes of -0.45 and -0.46 hours respectively, are -1.07 to +0.17 (20 mg/day) and -1.08 to +0.16 (40 mg/day) so that similar comments could be made at the separate dose levels. This also applies to the ON time without troublesome dyskinesia outcomes, based on an assumed magnitude of the true treatment effect of 0.41 hours (random effects meta-analysis for 20 mg/day + 40 mg/day); predicted range, -0.21 to +1.03 hours.

In order to exclude the potential for specific factors to have negatively influenced the primary outcome, leading to a non-significant result, the applicant undertook a systematic review of all istradefylline pivotal studies, including Study 6002-014. Study factors (including design, statistical methods, and study methods), site factors (including region, enrolment patterns, experience, and training) and patient factors (including disease characteristics, demographics, and concomitant medications) were all reviewed in detail. The review did not provide any evidence that specific factors directly contributed to the failure to meet the primary endpoint.

Evidence of Effectiveness in Study 6002-014

Although statistically significant separation was not seen for the pre-defined primary and secondary efficacy endpoints in Study 6002-014, analysis of the results, taking into account data collected throughout the duration of study treatment, did provide supportive evidence of a positive drug effect in maximally and optimally treated patients.

Justification for Additional Analyses

Given the variability in symptom manifestations, disease status, and daily burden in PD, it is often difficult to capture treatment benefit when evaluating response to treatment at a single timepoint within a study due to increased risk of capturing random data anomalies or other effects that result from the intrapatient variability noted above. Evaluating efficacy by taking account of available data at all timepoints during the study helps to address the inherent variation and represents a way of addressing the limitations associated with evaluating outcomes at a single assessment timepoint. These analyses provide additional insight into the treatment effect and the benefit to the patient.

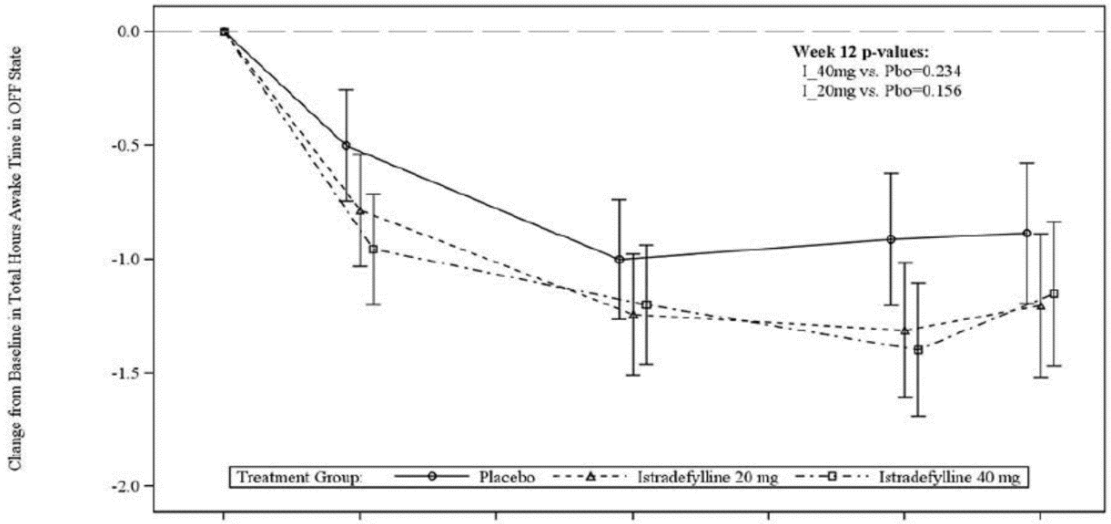
Regarding these analyses the applicant utilized 2 approaches; (i) analysis of LS means change from baseline data at each study visit for individual study treatments arms, and (ii) analysis of summary data collected across all visits using a single measure e.g., area under the effect curve (AUEC) analyses, which are discussed below.

LS means Change from Baseline at Each Study Visit

Figure 50 shows that for both doses of istradefylline, at each timepoint, numerically greater changes from baseline in OFF time were seen compared to placebo; the evident separation between plots providing support for a positive effect of istradefylline in this population.

The reversal seen in the change from baseline in time spent in the OFF state for both istradefylline treatment arms but particularly for the 40 mg/day arm at the last study visit, marks an unexpected shift in the downward trend up until that point and has been observed in other studies in the istradefylline program. As discussed above, these patterns in the data may occur as a result of heterogeneity in PD and exemplify the limitations of assessing benefit based on outcome at a single timepoint.

Figure 50: LS Mean Change from Baseline to Week 12 in Hours/day in the OFF State in Study 6002-014 (20 mg/day, 40 mg/day, placebo)



No. of Subjects					
Placebo	198	198	192	187	188
Istradefylline 20 mg	194	193	184	179	179
Istradefylline 40 mg	200	200	190	181	176

Source: 6002-014 CSR, Figure 14.2.1.1.1.

Time-Adjusted Area Under the Effect Curve Analyses

The AUEC analysis takes into account all recorded data post-baseline, with Week 2 representing the first study visit and the earliest timepoint at which change from baseline could be calculated. The AUEC for a particular outcome (e.g., change from baseline in time spent in hours in the OFF state) is standardized by dividing by the duration of time under consideration at the patient level (i.e., 10 weeks; Week 2 to Week 12); therefore, the units for the time-adjusted AUEC measure are hours. Post-hoc time-adjusted AUEC analyses for Week 2 to Week 12 were conducted in terms of the difference in LS means and corresponding 95% CI values from paired comparison of each istradefylline treatment arm versus placebo using an ANCOVA model.

Data on the 20 mg/day and 40 mg/day doses were combined for these analyses. This pooling increases the precision of the estimate and is considered appropriate to establish certainty of effect against placebo for both OFF time and ON time without troublesome dyskinesia outcomes. This is further explained in Ground 1.

Results of AUEC analysis of change from baseline in OFF time and ON time without troublesome dyskinesia for Study 6002-014 for the combined 20 mg/day and 40 mg/day doses are summarized in Table 137.

Table 137: Time-Adjusted AUEC for Change from Baseline in OFF State and ON State Without Troublesome Dyskinesia from Weeks 2 to 12 Compared with Placebo, for Study 6004-014 (ITT Analysis Set)

	Istradefylline 20 mg/day + 40 mg/day vs. placebo
OFF time (hours/day)	
Difference in LS means (95% CI)	-0.34 (-0.63, -0.05)
Nominal p-value	0.022
ON time without Troublesome Dyskinesia (hours/day)	
Difference in LS means (95% CI)	0.08 (-0.29, 0.44)
Nominal p-value	0.688

CI = confidence interval; ITT = intent to treat; LS = least squares.
Note: Baseline parameter value, study, study center nested within study, and treatment group are variables in the model; the analyses used multiple imputation (copy reference [CR]) under missing not at random assumptions.
Source: Module 5, Re-exam Table 15.1 and Re-exam Table 15.2.

Subjects treated with istradefylline 20 mg/day+40 mg/day combined experienced a nominally significant reduction versus placebo in daily time spent in the OFF state and a numerically greater increase versus placebo in time spent in the ON state without troublesome dyskinesia with istradefylline when assessed using the AUEC analysis of data for Week 2 to Week 12.

Despite a statistically significant treatment effect not being shown for the OFF time primary endpoint in Study 6002-014, a nominally significant difference was seen in the AUEC analysis of the change from baseline in hours OFF time outcome, which took account of the totality of the OFF time data i.e., data from both dose groups and at all post-baseline study visits. Results for the ON time without troublesome dyskinesia AUEC outcome presented above, also supported the presence of a positive effect of istradefylline in the study. It should also be noted that the additional benefits observed with istradefylline specifically in Study 6002-014 were accrued on top of maximal treatment with dopaminergic agents. These data indicate that despite the formal primary and secondary endpoint results, Study 6002-014 cannot be considered a study without any evidence of a genuine drug effect or positive results.

Efficacy in Maximally Treated Patients

In order to address CHMP's concerns regarding the efficacy of istradefylline in patients that could be described as 'maximally and optimally' treated, given the results of Study 6002-014, the applicant conducted analyses on the change from baseline for OFF time and ON time without troublesome dyskinesia outcomes measured in subjects across the 8 pivotal studies who met the 'maximally and optimally treated' criteria agreed with the FDA. The aim of the analyses was to determine whether the 3 critical characteristics adversely impacted the outcome of Study 6002-014.

As presented during the evaluation phase prior to the CHMP opinion on 22 July 2021, the analyses were expanded from Study 6002-014 to encompass subjects in the other pivotal studies who also met the 3 eligibility criteria distinct to Study 6002-014. Results of the istradefylline 20 mg/day and 40 mg/day treatment arms have now been pooled in order to provide more precise estimates of effects (Table 138). Justification for pooling is outlined in Ground 1.

In these analyses, 1213 subjects (placebo: 449 subjects [45.3% of ITT placebo subjects]; istradefylline 20 mg/day+40 mg/day: 764 subjects [44.2% of ITT subjects on 20 mg/day+40 mg/day combined

doses]) met the 3 key inclusion criteria for Study 6002-014 and 1506 subjects (placebo: 543 subjects [54.7%]; istradefylline 20 mg/day+40 mg/day combined doses: 963 subjects [55.8% of ITT subjects on 20 mg/day+40 mg/day combined doses]) did not.

All subjects enrolled in Study 6002-014 had a documented history of levodopa-induced dyskinesia. This information was not collected systematically in the other pivotal studies. However, the presence of dyskinesia was captured at baseline within a subject's diary and was systematically recorded across all 8 pivotal studies. Therefore, to allow direct comparison across the pivotal studies in the analyses of subjects stratified by presence of the 3 key Study 6002-014 eligibility criteria, confirmation of dyskinesia was based on the information recorded in the subject's baseline diary. For the purposes of this analysis therefore, 78.3% and 75.6% of subjects in the placebo and combined istradefylline 20 mg/day+40 mg/day groups, respectively, in Study 6002-014 are said to have met all 3 key inclusion criteria.

Table 138: LS Mean Change from Baseline at Week 12 in Total Hours OFF Time and Total Hours ON Time Without Troublesome Dyskinesia vs Placebo Based On Meeting the 3 Key Study 6002-014 Inclusion Criteria (Pool E1, ITT Analysis Set, OC, MMRM)

	Placebo	**Istradefylline 20 mg/day and 40 mg/day**
OFF time (hours/day)		
Subjects who **met** the 3 key entry criteria defined in Study 6002-014		
N	449	764
LS mean change (95% CI)	-0.70 (-0.93, -0.46)	-1.03 (-1.22, -0.84)
Difference in LS means[a] (95% CI)	-	-0.33 (-0.62, -0.05)
Subjects who **did not meet** the 3 key entry criteria defined in Study 6002-014		
N	543	963
LS mean change (95% CI)	-0.92 (-1.15, -0.68)	-1.35 (-1.53, -1.17)
Difference in LS means[a] (95% CI)	-	-0.43 (-0.71, -0.15)
ON time without troublesome dyskinesia (hours/day)		
Subjects who **met** the 3 key entry criteria defined in Study 6002-014		
N	449	764
LS mean change (95% CI)	0.67 (0.39, 0.96)	0.96 (0.73, 1.19)
Difference in LS means[a] (95% CI)		0.29 (-0.06, 0.63)
Subjects who **did not meet** the 3 key entry criteria defined in Study 6002-014		
N	543	963
LS mean change (95% CI)	0.76 (0.51, 1.00)	1.13 (0.94, 1.32)
Difference in LS means[a] (95% CI)	-	0.38 (0.09, 0.67)

CI = confidence interval; LS = least squares.
[a] Differences in LS means and corresponding 95% CI are from paired comparisons between each istradefylline treatment arm vs placebo. A MMRM approach was used with baseline assessment as a covariate, and study, study center nested within study, treatment group, week, and treatment-by-week interaction as fixed effect terms.
Pool E1 includes Studies 6002-US-005, 6002-US-006, 6002-US-013, 6002-0608, 6002-009, 6002-US-018, 6002-EU-007, and 6002-014.
Source: Module 5, Re-exam Table 6.1 and Module 5, Re-exam Table 6.2.

Comparison of OFF time and ON time without troublesome dyskinesia treatment effects in Pool E1 subjects (i.e., including those from Study 6002-014) who met or did not meet the 3 key Study 6002-014 inclusion criteria showed no substantive differences. Furthermore, the treatment effects in both

subgroups for the OFF-time endpoint were found to be nominally significant. For the ON time without troublesome dyskinesia outcome, a significant difference was observed in the subgroup not characterized by the 3 Study 6002-014 criteria. The result in the Study 6002-014-like subgroup narrowly misses statistical significance but nonetheless shows a strong positive trend that is consistent with the subgroup not characterized by the 3 Study 6002-014 criteria. Overall, the data showing the positive effects of istradefylline in the subject population with a higher dopaminergic load are in line with those of the complementary subgroup and of the overall Pool E1 cohort, supporting efficacy in this patient population.

Lastly, to further explore the effect of Study 6002-014 eligibility criteria on treatment outcome, the applicant assessed treatment × covariate interactions using an MMRM analysis across the 2 subgroups of subjects positive and negative for the 3 key Study 6002-014 eligibility criteria. The results showed that for both istradefylline 20 mg/day and 40 mg/day, there was no evidence of effect modification (i.e., these 3 factors in combination do not significantly influence treatment effect). Treatment × covariate interactions were not significant with p-values for OFF time (p=0.614 for istradefylline 20 mg/day and p=0.436 for istradefylline 40 mg/day) and ON time without troublesome dyskinesia (p=0.585 and p=0.500).

Based on the data presented within this section, the applicant asserts that the efficacy of istradefylline in those who meet the Study 6002-014 key inclusion criteria used to indicate maximal and optimal treatment does not differ from efficacy in those who do not meet the criteria. The evidence indicates that the population characteristics of the Study 6002-014 subjects did not contribute to the study not meeting its primary endpoint.

Evidence of Efficacy in the European Population

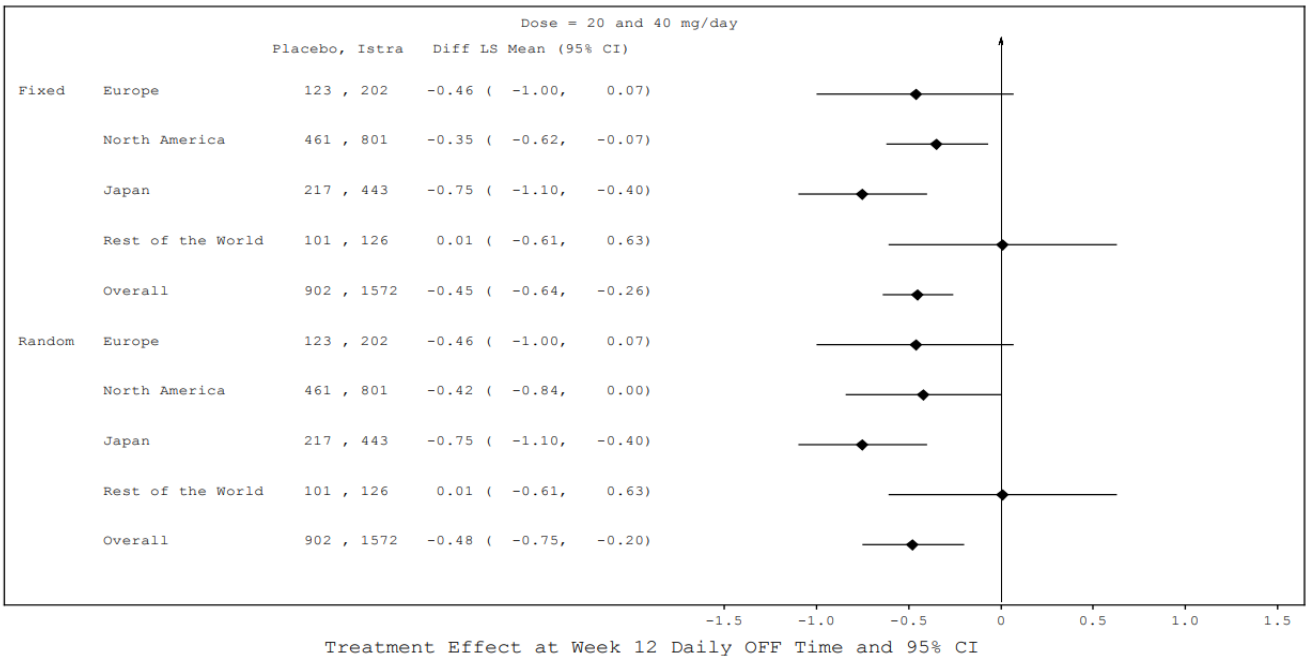
In order to address CHMP concerns around the efficacy of istradefylline in the European population and explore whether Study 6002-014 did not meet its primary endpoint due to inclusion of European subjects, the applicant conducted a number of regional subgroup analyses.

European subjects in the istradefylline pivotal study program were included in Studies 6002-EU-007 and 6002-014, which enrolled subjects from sites in Europe, Asia, South America, South Africa and North America. European subjects were enrolled to sites in Austria, Estonia, France, Italy, Latvia, Lithuania, Spain, the Ukraine and the UK (Study 6002-EU-007) and from the Czech Republic, Germany, Italy, Poland and Serbia (Study 6002-014). A total of 136 of 464 randomized (29.3%) subjects in Study 6002-EU-007 and 257 of 613 (41.9%) randomized subjects in Study 6002-014 constituted the ITT population at European sites. Studies 6002-014 and 6002-EU-007 were the only 2 studies in the pivotal study program that included European sites.

As shown in the responses provided during the evaluation phase prior to the CHMP opinion on 22 July 2021, the treatment effect assessed by random effects meta-analysis of LS mean difference in change from baseline at Week 12 in total daily hours of OFF time between istradefylline and placebo in European subjects (20 mg/day: -0.49 hours; 40 mg/day: -0.46 hours) was consistent with findings for the North American (20 mg/day: -0.30 hours; 40 mg/day: -0.33 hours) and overall subject populations (20 mg/day: -0.45 hours; 40 mg/day: -0.46 hours) for both istradefylline doses.

An overview of the results for the change from baseline to Week 12 in total daily hours of OFF time and ON time without troublesome dyskinesia analyses per region for istradefylline (20 mg/day and 40 mg/day combined compared to placebo) across Pool E1 (i.e., all 8 istradefylline pivotal studies) are presented in Figure 52 and Figure 53, respectively. The data from the istradefylline 20 mg/day and 40 mg/day treatment arms have been combined for the reasons outlined in Ground 1. The results for istradefylline 20 mg/day and 40 mg/day (i.e., separately) versus placebo for both OFF time and ON time without troublesome dyskinesia outcomes are also presented.

Figure 51: Meta-Analysis: LS Mean Difference from Placebo in the Change from Baseline to Week 12 Hours/day in the OFF State [95%CI] for Istradefylline (20 and 40 mg/day Combined) by Regional Subgroup - (OC, ITT, MMRM, Pool E1)

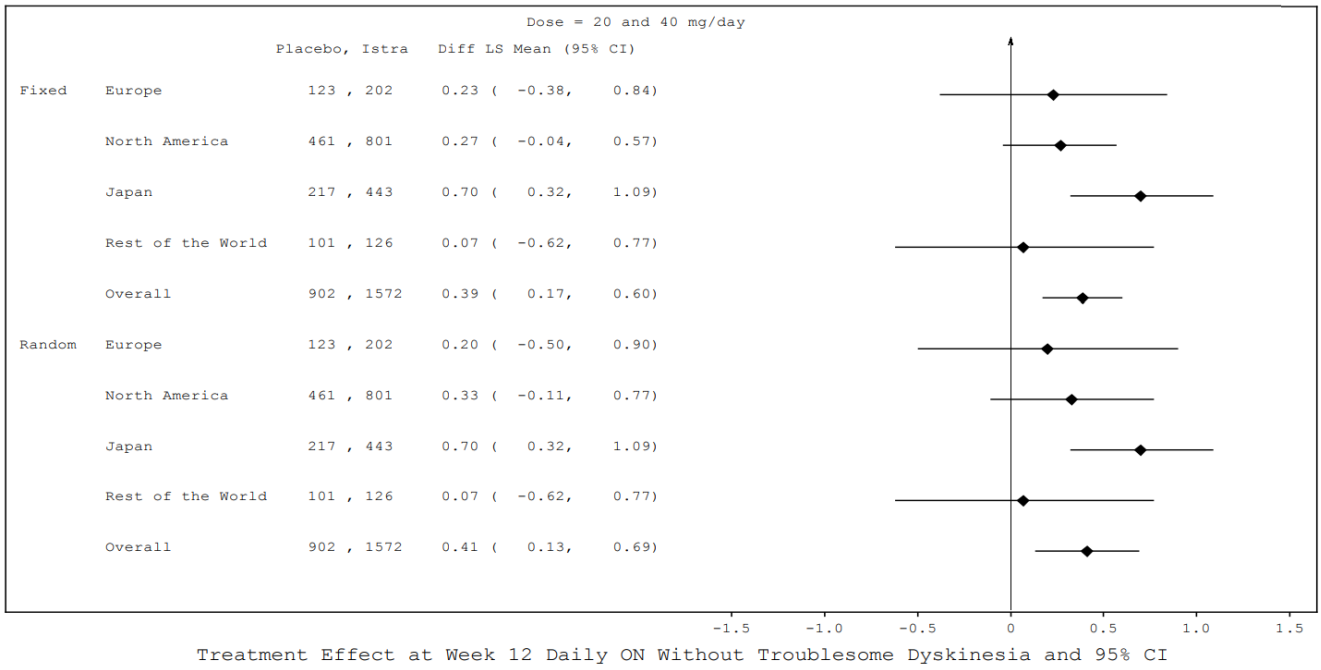


Source: Module 5, Re-exam Figure 2.1.2.

The treatment effect as assessed by random effects meta-analysis of LS mean difference in change from baseline at Week 12 in total daily hours of OFF time between istradefylline and placebo in European subjects (istradefylline 20 + 40 mg/day: -0.46 hours) was consistent with findings for the North American (istradefylline 20 + 40 mg/day: -0.42 hours) and overall subject populations (istradefylline 20 + 40 mg/day: -0.48 hours).

A positive numerical effect was also observed for the secondary endpoint (change from baseline at Week 12 in total daily hours of ON time without troublesome dyskinesia) in the European population.

Figure 52: Meta-Analysis: LS Mean Difference from Placebo in the Change from Baseline to Week 12 Hours/day in the ON state Without Troublesome Dyskinesia [95%CI] for Istradefylline (20 and 40 mg/day Combined) by Regional Subgroup - (OC, ITT, MMRM, Pool E1)



Source: Module 5, Re-exam Figure 2.2.2.

The result in the European subgroup (istradefylline 20 mg/day + 40 mg/day: 0.20 hours) was in line with findings in the North American subgroup (istradefylline 20 + 40 mg/day: 0.33 hours).

Positive treatment differences between istradefylline and placebo were observed for the LS mean change from baseline at Week 12 in OFF time and ON time without troublesome dyskinesia endpoints evaluated in the European population for the combined 20 mg/day and 40 mg/day dose groups, with the OFF-time treatment difference narrowly missing nominal statistical significance. It should be noted that the meta-analyses by region were not expected to show significant differences for the European subgroup as they were not powered for this outcome and as these were not pre-defined targets for confirmatory evaluation. The magnitude of the OFF-time treatment effect by point estimate was nonetheless similar to the significant random effects meta-analyses results for Pool E1 and therefore was supportive of a real treatment effect in the European population.

Given that European subjects in the istradefylline program were ultimately enrolled in 2 studies (both conducted across multiple regions) where active treatments formally failed to separate significantly from placebo for the primary endpoint, it is noteworthy that treatment effects of similar magnitudes to those in other regions and in the Pool E1 population were observed in the European cohort.

Generalizability of Data to the European Population

CHMP raised concerns regarding the generalizability of the positive findings from non-EU territories and overall in the istradefylline pivotal study program to the European population. To address these concerns, the applicant outlines below data showing the OFF time and ON time without troublesome dyskinesia treatment effects in the European subgroup to be consistent with those from other regions and overall, particularly once imbalances in baseline factors are taken into account. The lack of differences in intrinsic and extrinsic factors and the modest differences in approaches to therapeutic intervention in PD across

the regions indicate that differences in effectiveness of PD treatments from region to region are not expected.

Similarity of Istradefylline Effect Across Regions

Modest differences in OFF and ON time without troublesome dyskinesia outcomes were observed across regional subgroups, with slightly larger treatment effects seen in the Japanese studies when compared to studies enrolling subjects in other regions. As differences in baseline factors were seen across regions (, it was assessed whether any identifiable imbalances in demographic parameters, possible predictors of disease severity, baseline levodopa dose, concomitant anti-PD medications and other prognostic factors could influence OFF and ON time without troublesome dyskinesia treatment effects. Subject demographics and baseline characteristics by region are provided as part of the grounds of re-examination (data not shown in the CHMP AR).

Analysis of treatment × covariate interactions demonstrated that most of these baseline parameters did not impact on the efficacy at the Pool E1 or regional levels (treatment × covariate interaction, $p<0.1$).

Of those that were noted to have a statistically significant interaction test, only gender, total hours per day spent in the OFF state at baseline (baseline OFF time) and age (40 mg/day) remained as independent effect modifiers following multivariate regression analysis of the Pool E1 population. Of these, baseline imbalances across the regions were noted only for the gender and baseline OFF time outcomes and therefore considered to be relevant to this discussion.

Gender Effect

The significant treatment × gender interaction results from the finding that the OFF time treatment effect in females exceeds that for males (Table 139).

Table 139: LS Mean Difference from Placebo in the Change from Baseline to Week 12 Hours/day in the OFF state [95% CI] for Istradefylline 20 mg/day and 40 mg/day by Gender (OC, ITT, MMRM, Pool E1)

Population		Difference LS means (95% CI), 20 mg istradefylline	p-value for interaction*	Difference LS means (95% CI), 40 mg istradefylline	p-value for interaction*
Overall	Female (n=1035)	-0.67 (-1.03, -0.31)	P=0.066	-0.77 (-1.13, -0.42)	P=0.031
	Male (n=1439)	-0.18 (-0.50, 0.13)		-0.25 (-0.57, 0.06)	

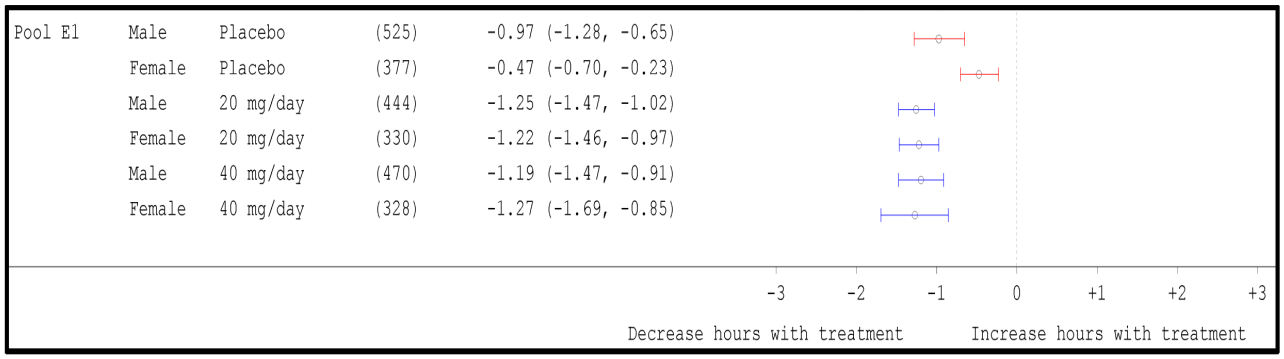
*p value for treatment x covariate interaction not for istradefylline vs placebo
Source: Module 5, Day 120 Table 2.9.1.1.1.

As noted by the applicant, multivariate regression confirmed gender to have an independent effect on the treatment difference (p=0.0649 (20 mg/day) and p=0.1030 (40 mg/day) in the final model).

Given that the Japanese studies enrolled a higher proportion of females (56.8%) than the North American (35.5%) and European (41.5%) studies, this is considered to contribute to the differential effect between Japan and European/North American subgroups.

As described in detail in responses provided during the evaluation prior CHMP opinion on 22 July 2021, the higher magnitude of response observed in females was driven mainly by a lower response in the placebo group, rather than a higher istradefylline response. In absolute terms, the responses to istradefylline were almost identical between men and women. This can be seen in Figure 53.

Figure 53: LS Mean Change (95% CI) from Baseline at Week 12 for Hours per Day Spent in the OFF State for Each Treatment Group by Gender (Random Effects Meta-Analysis, Pool E1, MMRM, ITT)



CI = confidence interval; ITT = intent-to-treat; OC = observed case.
Pool E1: Studies 6002-US-005, 6002-US-006, 6002-US-013, 6002-US-018, 6002-EU-007, 6002-009, 6002-0608 and 6002-014
Source: Module 5, Day 180 Figure 9.1.1.

The causes of the gender influence on the placebo effect seen in the istradefylline pivotal study program are unclear. The critical finding however, is the consistent absolute response across all istradefylline treatment arms by gender.

Overall, the data above show that a numerical benefit for istradefylline is achieved for both females and males, with females estimated as having a larger treatment effect than males. Given that a greater proportion of females were recruited into the Japanese studies, the applicant considers that when gender is taken into account, the treatment effects in Japan and Europe/North America are more closely aligned.

Baseline OFF Time Effect

A statistically significant treatment x covariate interaction effect (istradefylline 20 mg/day: p=0.0023; istradefylline 40 mg/day: p=0.0165) was seen for total hours per day in the OFF state at baseline at both dose levels overall, with subjects with more OFF time at baseline experiencing a larger treatment benefit from istradefylline. Multivariate regression indicates this being an independent effect (p=0.0064 (20 mg/day) and p=0.0097 (40 mg/day) in the final model). The influence of total baseline OFF time on the magnitude of change from baseline in OFF-time treatment effect is further discussed in Ground 1.

The baseline total hours per day spent in the OFF state in the European population (5.63 hours) was lower than in the Japanese population (6.43 hours). Given the strength of the interaction, this difference in baseline OFF time further contributes to the observed treatment effect differences between the Japanese and European populations.

Generalizability of Data to European Population

ICH guideline E5 (Ethnic Factors in the Acceptability of Foreign Clinical Data) was reviewed to determine to what extent data from istradefylline studies in North America and Japan can be used to support the approval of istradefylline in the EU; the key points are summarized below:

- The enduring principles of PD diagnosis, levodopa-based treatment, and treatment of motor fluctuations are similar across Europe, North America and Japan. European, Japanese and US guidelines recommend that wearing-off should be managed by adjusting the dose/formulation of levodopa, or via use of adjunctive pharmacotherapy. The same PD adjunct treatments are in use across these territories; with the exception of zonisamide, which is approved for treatment of PD in Japan but not in USA or Europe and istradefylline, which is approved in USA and Japan.
- The slight differences across regions in approaches to use of pharmacotherapy in treating PD were included in the grounds for re-examination. Note that a detailed analysis of treatment ×

covariate interactions and multivariate regression analyses did not demonstrate a significant treatment interaction related to concomitant medications use or levodopa dose administered. The regional clinical practice differences outlined above are therefore not considered to impact the generalizability of data to the EU population.

- PopPK analyses demonstrated that istradefylline PK factors of concomitant CYP3A inhibitor use and smoking had meaningful effects on exposures. While race was found to be a statistically significant covariate with effects on clearance, the effects are not considered clinically meaningful, i.e., non-Asian and Japanese patients have comparable exposures.
- Exposure-response analyses showed that over the 20 mg to 60 mg dose range there was no apparent relationship between istradefylline exposure and the primary clinical endpoint (change from baseline in total OFF hours per day) or the incidence of the most frequent AEs. Any differences in istradefylline exposure between Japanese and non-Japanese patients were therefore not clinically relevant.
- There are not believed to be any differences in the pathophysiology of PD across different regions, specifically no reason to think that there might be relevant pharmacogenomic differences.

Data from the European population showed that istradefylline was associated with a positive treatment effect, which was generally considered to be of similar magnitude to the effect observed in other regions, once differences in baseline factors (mainly gender and total daily hours spent in the OFF state) were taken into account. The consistency of principles of therapeutic treatment and lack of significant differences in important intrinsic and extrinsic factors across regions indicate that the regional variations in istradefylline treatment effect are not a function of race or region.

Overall, the evidence indicates that the data from across the regions in the istradefylline pivotal study program are generalizable to the European population.

Change in Treatment Practices Over Time

The standard of care of medical interventions for PD patients experiencing end-of-dose wearing off and eligible for further orally administered therapy has changed little over the last few decades and over the course of the istradefylline development program, with few differences between regions. Replacement of dopamine or modulation of dopaminergic activity remains the mainstay of therapy for PD (Jimenez-Shahed, 2016) and has been since the 1970s.

Despite the continued use of dopaminergic therapy and the number of approved therapies available, there is no established pathway and little structure regarding the use of these agents in clinical practice; though suggested algorithms for use do appear in some published guidelines. Globally, the treatment approach to this day has remained one of empirical medicine (‘trial and error’), largely due to the heterogeneity and unpredictability of response to treatment, as exposed by the applicant during the evaluation phase prior CHMP opinion on 22 July 2021.

The lack of new therapies with novel mechanisms of action and the persistence of a trial and error approach to treatment, suggest that for PD patients who are eligible for further orally administered therapy and currently experience end-of-dose wearing off, the treatment landscape has not changed for the last few decades and since the start of the istradefylline pivotal study program. Consequently, the applicant considers that the patients treated in the pivotal studies are still representative of the contemporary European population who would be eligible for istradefylline in event of MA approval and that similar responses to istradefylline treatment would be expected.

Summary and Conclusions

Despite the formal primary and secondary endpoint results, the data in Study 6002-014 show that istradefylline was associated with clearly positive trends in the treatment effects across both primary and secondary endpoints, at both 20 mg/day and 40 mg/day dose levels.

The magnitude of the OFF time and ON time without troublesome dyskinesia treatment effects have been shown to fall within the predicted range when the estimated istradefylline treatment effect, sample size per treatment arm and interpatient variability are taken into account. Notably, the results of the time-adjusted AUEC analyses (of Study 6002-014), which address the limitations of evaluating the treatment effect at a single timepoint, caused by the marked inter- and intrapatient variability in PD, show nominal statistically significant differences versus placebo for the primary efficacy outcome. Taken together, these data indicate that Study 6002-014 cannot be considered a study without any evidence of a genuine drug effect or positive results.

To address concerns that subject characteristics, specifically the recruitment of 'maximally and optimally' treated patients (Study 6002-014 only) and European patients (Studies 6002-EU-007 and 6002-014) to the studies, may have led to these studies failing to show significant differences for their primary endpoints, further subgroup analyses by region and in the population meeting the main eligibility criteria for Study 6002-014 were undertaken and showed that neither factor was a key contributor to the observed treatment effect size.

It should be noted that there are no intrinsic and extrinsic differences between the Japanese and European populations that suggest potential differences in response to treatment at the patient level. Given the above and the distinct lack of new therapeutic classes of PD adjuncts and persistence of the empirical approach to PD treatment over the last 2 decades, the applicant considers that the data from the istradefylline program adequately reflects the anticipated response to treatment in the present-day EU population.

Overall, the data provide support for a positive effect of istradefylline in Study 6002-014 and indicate that the treatment effects seen in the overall population are representative of those expected in 'maximally and optimally' treated patients and in European patients.

CHMP discussion on ground for re-examination #2

Study 6002-014 was conducted at the request of the US FDA to study istradefylline in PD patients who were considered to be 'maximally and optimally treated' (i.e., a population with a higher dopaminergic load than the earlier istradefylline studies). It also enrolled European patients. As such it is considered more important than the other studies in the dossier. However, it did not provide positive results for the primary endpoint (OFF time), nor for key secondary endpoints (ON time and UPDRS Part III).

The applicant argues that the results of the 014 study falls within the predicted range. This may be true, but the fact is that all eight studies fall within (or are close to) this predicted range, because the range was based on the same eight studies. Furthermore, four of these studies did not reach statistical significance. Roughly speaking, this means half of the predicted range does not cover effect sizes that are statistically significant let alone are considered clinically relevant.

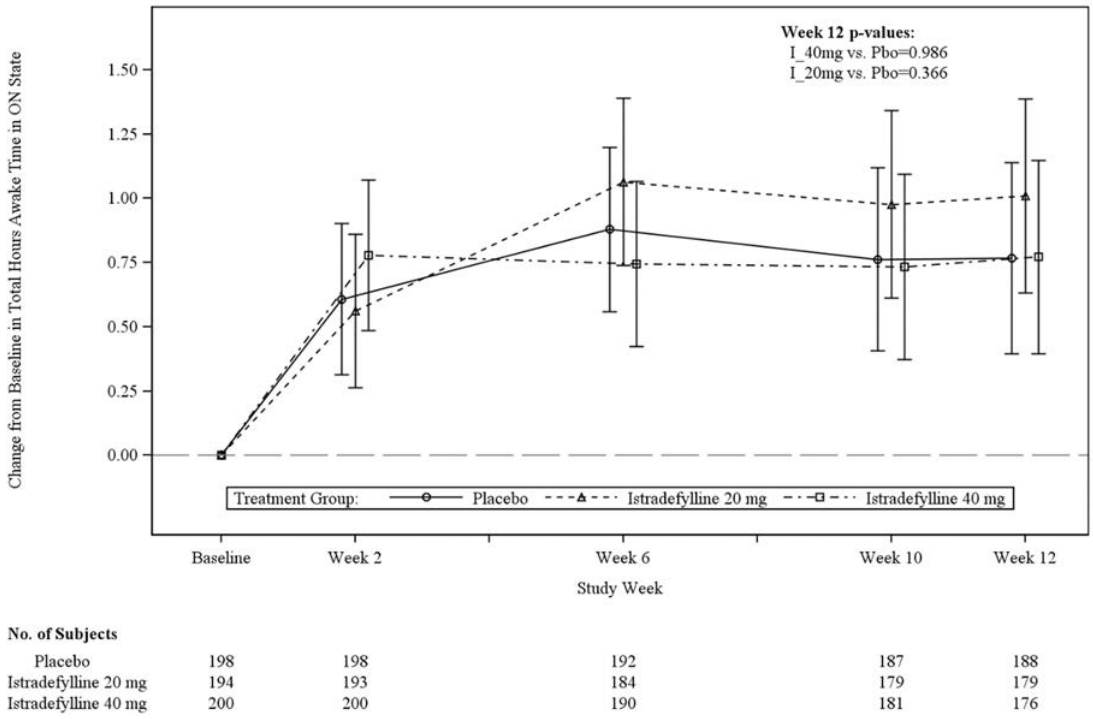
The applicant continues with several *post-hoc* analyses, which are considered exploratory.

In a plot of hours in OFF time over time (Figure 50), the applicant concludes that a reversal of effect is seen towards the end of the trial, an unexpected shift in the downward trend up until that point. However, using the same argumentation, it can also be that the week 10 results are unexpectedly low and the aberrant one. This is not convincing evidence that this study provided positive evidence of efficacy.

The time-adjusted AUEC analysis would take into account all data post-baseline. Here pooling of doses is needed to reach nominal statistical significance, this is not acceptable. As already explained in the assessment of Ground #1, the original analysis of the study had separate analyses for the two doses, this would be expected in post-hoc analyses as well. Results for the individual doses were not provided. Moreover, there is clearly no effect on the secondary endpoint (ON time) and a very small effect, hardly diverging from zero for the primary endpoint (OFF time).

This uncertainty is not dissipated when the daily time in the ON state without troublesome dyskinesia was considered. Neither each study visit approach (see figure below) nor the AUEC analysis (difference in LS Means [95%CI] = 0.08 [-0.29, 0.44]) provided confirmation of the observed (modest) effect on the time spent in the OFF state.

Figure 54: Key Secondary Efficacy Endpoint: Change from Baseline (LS Mean with 95% CI) of Awake Time/Day in the ON State without Troublesome Dyskinesia

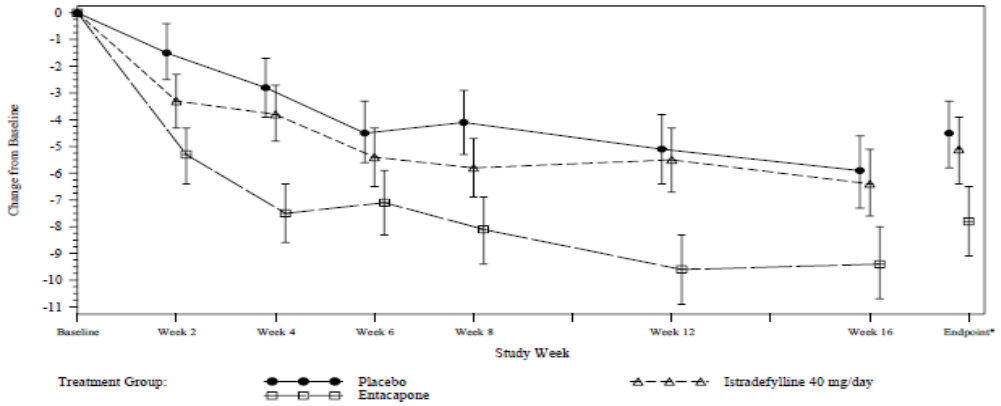


No. of Subjects					
Placebo	198	198	192	187	188
Istradefylline 20 mg	194	193	184	179	179
Istradefylline 40 mg	200	200	190	181	176

Source: 6002-014 CSR; Figure 14.2.2.1.1

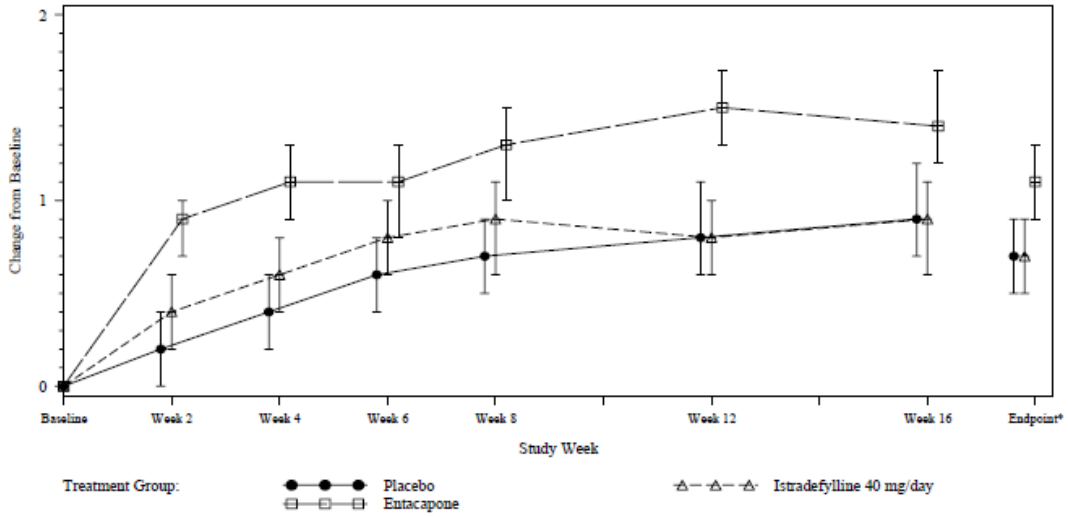
While not discussed by the applicant in the responses document, it is worth mentioning that a similar pattern was also observed in the second EU study (Study 6002-007). It was shown for daily time spent in the OFF state, while not for the istradefylline arm. There was an evident separation with respect to placebo for the active control arm entacapone (Figure 55).

Figure 55: Primary efficacy: change from baseline by study visit at endpoint in the percentage of awake time per day spent in the OFF state-observed-case analysis (Intent-to-Treat analysis set)



No support was also obtained from the analysis of the secondary endpoint: Change from Baseline (LS Mean with 95% CI) of Awake Time/Day in the ON State without Troublesome Dyskinesia (Figure 56).

Figure 56: Change from baseline (LS Mean +/- SE) in the total hours of awake time per day spent in the ON state without troublesome dyskinesia. Based on patients´s ON/OFF Diary by Study visit – Observed Case Analysis ITT Analysis set



Pooled analysis of maximally treated patients (according to the inclusion criteria of study 6002-014), again uses pooling of the two doses to reach statistical significance. As this is not according to the SAPs of the individual studies where separate analyses for the two dose arms are defined. Furthermore, history of levodopa-induced dyskinesia was not recorded systematically in all studies and the treatment effect was smaller in the maximally-treated population than in the not maximally treated population. This is not considered convincing.

It is not understood how a subgroup analysis of patients meeting or not meeting key inclusion criteria of Study 6002-014 can prove that Study 6002-014 results should be seen as positive despite them not reaching statistical significance. At most, it can show that the key inclusion criteria were likely not the reason for Study 6002-014 being negative.

Further, the applicant provided meta-analysis per region, showing that the effect on OFF and ON time in European subjects are of similar magnitude as for US subjects and the overall population. The fact

remains that no statistically significant effect has been observed in the individual studies that includes European patients, nor in a meta-analysis combining the European patients. The reasoning that the meta-analysis by region was not powered is not followed, as the individual studies included in the meta-analysis were powered and the total number of European subjects is in the same range as the sample size of the individual studies.

Results for the change in total daily hours of OFF time and ON time without troublesome dyskinesia analyses were assessed per region by random effects meta-analysis. The size of the different subgroups was not balanced, the largest being North America (n=1265 patient) followed by Japan (n=660) and Europe (n=325). Rest of the World comprised 226 patients. Istradefylline 20 mg and 40 mg doses were combined.

Apart from the consistent effect observed in Japanese patients (see Fig 7 above) no clear conclusion can be drawn for the remaining regions, which showed a smaller and more variable effect size. The overall measured effect on OFF time (Diff LS Mean [95% CI] -0.48 [-0.75, -0.20]) appears to be essentially driven by the Japanese subgroup. These conclusions are also valid for the effect observed on ON time without troublesome dyskinesia measurement.

Some differences between regions were detected at baseline; e.g Japanese subgroup showed a higher proportion of females in (56.8% vs 41.5% in Europe and 35.5% in North America), lower mean weight (54.9 kg vs 77.7 kg vs 80.05 kg, respectively), lower daily dosage of levodopa (422.8 mg vs 805 mg vs 780.8 mg, respectively), even after corrected by body weight, and higher proportion of concomitant use of MAO-B inhibitors (51.2% vs 16.6% vs 21.9%) among others. It is difficult to conclude whether any or all these factors have a direct impact on the observed discrepancies. The applicant have speculated that gender may play a role, leading to a higher treatment effect in females than in males. This has been attributed to a lower response to placebo. Whatever may be the explanation, the variability of the data and the difficulty of interpreting the differences observed between regions prevents from drawing a sound conclusion on this issue.

The inconsistency of the data and the lack of support from the European study 007 (also failed) makes the assessment of robustness difficult.

In conclusion, post-hoc analyses cannot be accepted as pivotal evidence upon inconsistent or negative results in the primary and secondary analyses. Furthermore, the post-hoc analyses results are also not convincing from a clinical point of view. Finally, the region analysis only increases the concerns regarding a potential effect of istradefylline in the European population.

Point not resolved.

5.1.3. Ground #3

Grounds for re-examination:

The applicant contends that there is a misunderstanding in the CHMP's interpretation of Study 6002-US-006, for which the primary endpoint is statistically significant by protocol defined analysis methods.

applicant's Position on Ground 3

- ANCOVA is recognized as the most appropriate analysis for the primary endpoint in Study 6002-US-006. This method of analysis was pre-specified in the protocol, but was specified in the Statistical Analysis Plan to be ANOVA.
- Use of ANCOVA leads to statistically significant differences at each of the 2 dose levels of istradefylline compared to placebo for the primary endpoint, ANOVA does not.

- A misinterpretation in the Clinical Study Report regarding the gatekeeper test has led CHMP to conclude that evidence for a treatment effect of istradefylline compared to placebo for the primary endpoint from the ANCOVA analysis is marginal. This is not correct.
- Study 6002-US-006 provides strong support for an effect of istradefylline compared to placebo in reducing the amount of daily OFF time (ANCOVA, p=0.026, 20 mg/day, p=0.024, 60 mg/day).

ANOVA versus ANCOVA

The pre-planned primary statistical analysis comparing the treatment means for the reduction in the percentage of awake time spent in the OFF state (endpoint minus baseline) in protocol amendment 3 (dated 26 Nov 2002) was ANCOVA with the percentage of awake time in the OFF state at baseline included as a covariate.

As detailed in Table 140, this primary analysis was changed in the SAP (dated 02 Oct 2003), before database lock and unblinding, to be based on ANOVA, with the ANCOVA analysis being supportive. The reasons for this change are not known and were made in the absence of any unblinding of the data. Database lock took place on 20 Nov 2003 and database unblinding took place on 30 Nov 2003.

Table 140: Analysis Methods Stated in the 6002-US-006 Clinical Study Protocol, Statistical Analysis Plan and Clinical Study Report

Document	Date	Primary analysis method	Supportive analysis method
Protocol Amendment 3	26 Nov 2002	ANCOVA	-
Statistical Analysis Plan	02 Oct 2003	ANOVA	ANCOVA
Database lock	20 Nov 2003	NA	NA
Database unblinding	30 Nov 2003	NA	NA
Clinical Study Report	02 Mar 2007	ANOVA	ANCOVA

ANCOVA = analysis of covariance; ANOVA = analysis of variance; NA = not applicable.

Despite the switch from ANCOVA to ANOVA as the pre-specified method of analysis in the Statistical Analysis Plan and as reported in the CSR, it is well recognized that ANCOVA is the more appropriate analysis in a setting where the endpoint is defined in terms of a change from baseline ('In randomized studies and studies with treatment assignment depending on the baseline, ANCOVA must be used'; Van Breukelen [2006]). ANCOVA is now regarded as the "gold standard" methodology. Such an analysis has 2 advantages over ANOVA:

1. It corrects for baseline imbalances in the percentage of awake time spent in the OFF state at baseline. This is essential to account for any bias resulting as a consequence of regression towards the mean.
2. It accounts for inter-subject variability in the percentage of awake time in the OFF state at baseline. This in turn leads to reductions in the standard errors for the estimates of treatment effect.

With respect to baseline imbalances, the mean baseline percentage of awake time spent in the OFF state was 36.56% in the placebo group compared to 34.81% and 35.07% in the istradefylline 20 and 60 mg/day groups, respectively. Failing to take account of these baseline imbalances in the analysis using ANOVA introduces bias into the comparison, whilst ANCOVA accounts for that imbalance. In other analyses that have been undertaken within this procedure, baseline OFF time has been seen to be a key driver of the subsequent reduction in OFF time from baseline, strengthening the position that ANCOVA is the most appropriate method of analysis. Statistical significance was achieved for both dose levels of istradefylline using the appropriate analysis method, ANCOVA.

Gatekeeper Analysis

The ANOVA test comparing the 3 treatment means was to act as a gatekeeper associated with a hierarchical testing procedure for subsequent evaluations of the individual dose levels versus placebo. If the ANOVA p-value for comparing the 3 treatment means was significant at the 5% level, statistical significance for the individual dose levels could be declared if both of those treatment comparisons gave $p<0.05$ (2-sided). This procedure would maintain the overall type I error rate at 5%. A similar procedure was to be adopted for ANCOVA as a supportive analysis according to the SAP.

As seen in Table 141, and based on an ANOVA methodology, statistical significance for the overall comparison of the 3 treatment groups was not achieved ($p = 0.169$). However, for the ANCOVA methodology, $p=0.049$ for the overall comparison allows testing to proceed with investigation of the individual istradefylline doses against placebo, and statistical significance was achieved with $p=0.026$ for istradefylline 20 mg/day, and $p=0.024$ for istradefylline 60 mg/day. These p-values are not marginal.

Table 141: Study 6002-US-006 Primary Efficacy: Change from Baseline in Percentage of Awake Time per Day Spent in an OFF State

	Percentage of Awake Time per Day Spent in an OFF State[a]			
Visit	Placebo (N=77)	Istradefylline 20mg/day (N=163)	Istradefylline 60mg/day (N=155)	Overall 2-sided p-value[b]
Baseline				
n	77	163	155	
Mean	36.56	34.81	35.07	
SD	14.336	14.701	13.748	
Median	36.90	32.40	35.10	
Min to max	4.2 to 81.4	8.2 to 82.5	6.2 to 72.4	
Endpoint				
N	75	162	148	
Mean	32.09	26.87	26.80	
SD	18.328	15.376	15.263	
Median	33.50	25.70	25.80	
Min to max	0.0 to 89.1	0.0 to 76.0	0.0 to 75.7	
Change from Baseline at Endpoint				
n	75	162	148	
Mean	-4.27	-7.94	-8.13	
SD	17.214	15.518	14.164	
Median	-3.90	-7.35	-8.65	
Min to max	-50.1 to 50.6	-67.9 to 32.7	-56.8 to 51.9	
ANOVA				0.169
LS mean	-4.07	-7.72	-7.84	
LS mean difference (versus placebo)		-3.65	-3.77	
p-value (versus placebo)[c]		0.088	0.082	
ANCOVA				0.049
LS mean	-3.47	-7.83	-7.96	
LS mean difference (versus placebo)		-4.36	-4.49	
p-value (versus placebo)[c]		0.026	0.024	

a Based on subjects' valid ON/OFF PD diary data from observed-case analysis.
b The overall p-value based on Type III Sums of squares F-test with 2 degrees of freedom for ANOVA or ANCOVA.
c P-value for individual comparisons (istradefylline groups versus placebo group) based on LS means from 2 way ANOVA, including terms for Investigator and treatment or ANCOVA, including terms for Baseline, Investigator, and treatment. At Endpoint, a closed testing procedure was used to test for treatment effects.
Source: 6002-US-006 CSR, Table 11.4.1-1.

In each of the above cases, these p-values derive from the gatekeeper analysis which was a simultaneous comparison of the 3 treatment means (istradefylline 20 mg/day, istradefylline 60 mg/day, and placebo). The following was the statement in the SAP: '*First, the overall test for treatment effects will be carried out using the F-test with two degrees of freedom from the Type III sums of squares in the ANOVA model described above using a significance level of 0.050*'. The 2 degrees of freedom test is a simultaneous test for all 3 means and is not a comparison of the 2 istradefylline doses (either individually or combined) versus placebo.

The wording in the CSR, which the CHMP referred to, was "*Based on the ANOVA model, the overall treatment effect compared to the placebo group was not statistically significant*". As described above, this statement should have referred to the gatekeeper analysis, and was mistakenly expressed in this way by the applicant, as it is not a comparison to the placebo group. This is therefore an error in the interpretation of the results in the CSR and this has been copied over into the CHMP assessment of the istradefylline application.

The p-values for the comparison of the combined istradefylline groups (20 mg/day and 60 mg/day) versus placebo in terms of LS mean differences were p=0.060 (ANOVA) and p=0.014 (ANCOVA).

Evidence of Efficacy

It has been stated by CHMP that the statistical significance for ANCOVA with a nominal p-value of 0.049 is marginal. As described above, the purpose of that overall analysis was to act as a gatekeeper, and it is the individual p-values of 0.026 (istradefylline 20 mg/day) and 0.024 (istradefylline 60 mg/day) that are the comparison of each dose versus placebo. These p values should be the focus when assessing the statistical significance of istradefylline, and they are not marginal as they are well below 0.05.

Summary of applicant's Position

Whilst is it acknowledged that the ANOVA was conducted based on pre-specification according to the SAP, failure to take the statistically significant results from the Protocol pre-specified ANCOVA methodology into account does not allow an unbiased and properly informed interpretation of the data for Study 6002-US-006 and its contribution to the overall benefit of istradefylline.

The applicant proposes that, irrespective of the final assigned status of Study 6002-US-006 by the CHMP on technical grounds (i.e., failed or not failed), due consideration should be given to the data in demonstrating statistical significance in pairwise treatment comparisons for each dose independently compared to placebo when the original protocol pre-specified analysis (ANCOVA) is applied. Further, the magnitude of the treatment effects are consistent with treatment effects demonstrated in other positive studies leading to the conclusion that istradefylline has shown efficacy in this study.

CHMP discussion on ground for re-examination #3

It is agreed with the applicant that ANCOVA is the preferred primary analysis for continues change from baseline endpoints. However, the fact remains that when the statistical analyses were worked out in the SAP of study 6002-US-006, the choice was made to use ANOVA instead. The applicant states the reason for this is not known, perhaps it was thought that randomisation would take care of baseline imbalances. The applicant intends to provide literature support for their perspective (Van Breukelen, 2006). This reference argues for the value of the ANCOVA with respect to more power, and while this is agreed such an argument cannot support a *post-hoc* change. On the contrary, this literature reference confirms the applicability of the ANOVA model, as being unbiased in the randomised setting. Therefore, the predefinition of ANOVA prevails and the ANCOVA remains a supportive analysis in this case and the study is formally negative. Furthermore, the switch in focus to ANCOVA is based on baseline imbalances and it is a fact that these have to been seen to be a key driver for subsequent changes. As such information

concerns *post-hoc* observations, this is a further argument to consider the ANCOVA analyses as exploratory.

Next, the applicant explained that the overall p-value in the ANOVA and ANCOVA was used for gatekeeping only and that interpretation of the results should focus on the p-values of the individual dose arms. The applicant correctly describes the testing principle and this is an acceptable method for handling multiplicity. They also correctly note that the p-value of 0.049 from the ANCOVA refers to the global F-Test and this has not been specified to that level of clarity previously. Further it is agreed that the focus should be on these p-values per dose arm. Thus, the p-values to consider for the exploratory ANCOVA analysis are 0.026 (20 mg/day) and 0.024 (60 mg/day) instead of 0.049. These p-values are well below 0.05 but the 95% confidence intervals are still considered wide (-8.16, -0.54 and -8.35, -0.62) with the lower limit close to zero. Nevertheless, with respect to the testing procedure this still means that the path to a conclusion of statistical significance for the 20mg dose still depends on the borderline passing of the gate and thereby the post-hoc change. The applicant's clarification does not change that Study US-006 remains to be considered as failed.

In conclusion, the fact remains that the predefined primary analysis, using ANOVA, failed to show an effect for either dose arm and the study is considered negative.

Point not resolved.

5.1.4. Additional ground for re-examination

Grounds for re-examination:

As a response to these grounds, the applicant submitted the grounds for re-examination including an additional ground discussing clinical relevance:

The applicant does not agree with comments in the assessment reports regarding the size of the treatment effect. The observed effect is relevant and clinically meaningful to patients with PD based on the feedback the applicant has received from patients surveyed, expert treating physicians as well as patient advocates and proposes that patients would benefit from the option of a novel, non dopaminergic treatment that offers a meaningful reduction in OFF time with a well characterized and known safety profile

Summary of applicant's Position on this ground for re-examination

This Ground for re-examination provides evidence from 3 separately conducted contemporaneous patient surveys outside of the istradefylline program and an exploration of cumulative benefit with increasing baseline OFF time from the istradefylline pivotal study program.

Istradefylline Treatment Effect

For reducing daily OFF time, a random-effects meta-analysis showed LS mean differences from placebo of -0.45 hours (95% CI: -0.75, -0.15), p=0.0031 for istradefylline 20 mg/day and -0.46 hours (95% CI: -0.80, -0.12), p=0.0074 for istradefylline 40 mg/day. This overall treatment effect equates to a *mean* 27-minute improvement per day in OFF time over placebo in the overall pivotal study population.

Patient Surveys, clinical Expert Statements and European PD Association Letter of Endorsement

The applicant sought to further understand the patient perspective regarding OFF and ON time improvement using 3 surveys:

A quantitative survey of European PD patients conducted and analyzed independently of the applicant by Adelphi Real World Research.

An online survey was conducted in patients with PD in Spain, Italy, France, Germany, and the UK. Eligible participants were those taking PD medications and experiencing OFF time. Questions 1 to 14 related to patient characteristics; Questions 15 to 22 related to the patient's perceptions of OFF time and ON time. Perceived benefit relating to improvements in OFF time and ON time were combined and categorized as: 'no difference', 'a little difference', 'moderate difference', 'quite a difference', and 'extreme difference'. The data were combined to create categories of 'no difference', 'a little difference', and 'moderate-extreme difference'.

In the survey, 101 participants (72% male; 65.2 mean age) were included. Thirty-four percent had a disease duration since diagnosis between 6-10 years and 24% had disease duration longer than 10 years. During a 24 hour period, 60% experienced a total OFF time of one hour or less and only 5% experienced a total OFF time of six or more hours.

The most relevant questions were Q19-Q22 (Table 142). Participants were asked "*If you were prescribed a new medication that could decrease OFF time by a total of 20 minutes throughout each day, with very few additional side effects, would this make a positive difference to you?*" Similar questions were formulated regarding a decrease in OFF time of 30 minutes and an increase in ON time without troubling dyskinesia of 20 minutes and 30 minutes.

Table 142: Results from questions 19-22 of the quantitative Adelphi Real World Research survey

	No difference at all	A little difference	Moderate-Extreme difference
20 minutes decrease OFF time	3%	19%	78%
30 minutes decrease OFF time	2%	20%	78%
20 minutes increase ON time without troubling dyskinesia	1%	23%	76%
30 minutes increase ON time without troubling dyskinesia	0%	25%	75%

Table built based on figures 10-13 provided in the grounds for re-examination

Additionally, subgroup analysis comparing those with ≤ 2hours OFF time versus those with > 2 hours OFF time was presented. A lower proportion of patients felt a decrease of 20 minutes in OFF time would be moderately-extremely beneficial (68%) in the group of > 2 hours OFF time compared to experiencing ≤ 2 hours OFF (82%). The difference between the two groups was less prominent for a 30 minutes OFF decrease (72% v 80%).

A qualitative survey of European patients living with PD and their caregivers, conducted and analyzed independently of the applicant by ThinkGen (April-May 2021)

Patients with PD and their family caregivers were recruited from France, Germany, Italy, Spain, and the UK. Participants were required to have a clinical diagnosis of PD and ongoing experience of OFF episodes or OFF time that impacted their daily routines and activities.

In the survey, 36 participants (53% male; 62.4 mean age) were included. Thirty-three percent had a disease duration since diagnosis between 6-10 years and 47% had disease duration longer than 10 years. During a 24 hour period, 59% experienced a total OFF time of one-two hours and only 6% experienced a total OFF time of five or more hours.

Most participants indicated that 20 minutes less OFF time would be highly beneficial and meaningful to their day-to-day lives, with quality of life implications across 3 broad categories of social, physical, and emotional benefits. When presented with the scenario of 30 minutes decreased OFF time, participants perceived greater benefit.

The Parkinson's Disease and Movement Disorder Alliance (PMDA), an independent, non-profit organization based in the US, provided online survey data to the applicant, who subsequently undertook independent analysis (February to March 2021)

The survey was conducted with respondents who either have PD or are a loved one of someone with PD and aimed to understand how a reduction in OFF time could improve quality of life. Analysis was based on results from 408 respondents, both carers and patients. Overall, 284 respondents were identified as patients (39.8% male; 73.6% 60-80 years) including 37.3% with disease duration below 5 years and 40.1% with disease duration between 5-10 years.

The first question asked the subject to rank thirteen activities from most important to least, in terms of what they would do if they had an additional hour when their medication was working. Responders were also asked to describe what they did in a recent 30-minute period when their medication was working. As this was a free text answer, only an exploratory, unvalidated analysis was able to be performed. The results from the US survey were consistent with findings from the 2 European surveys.

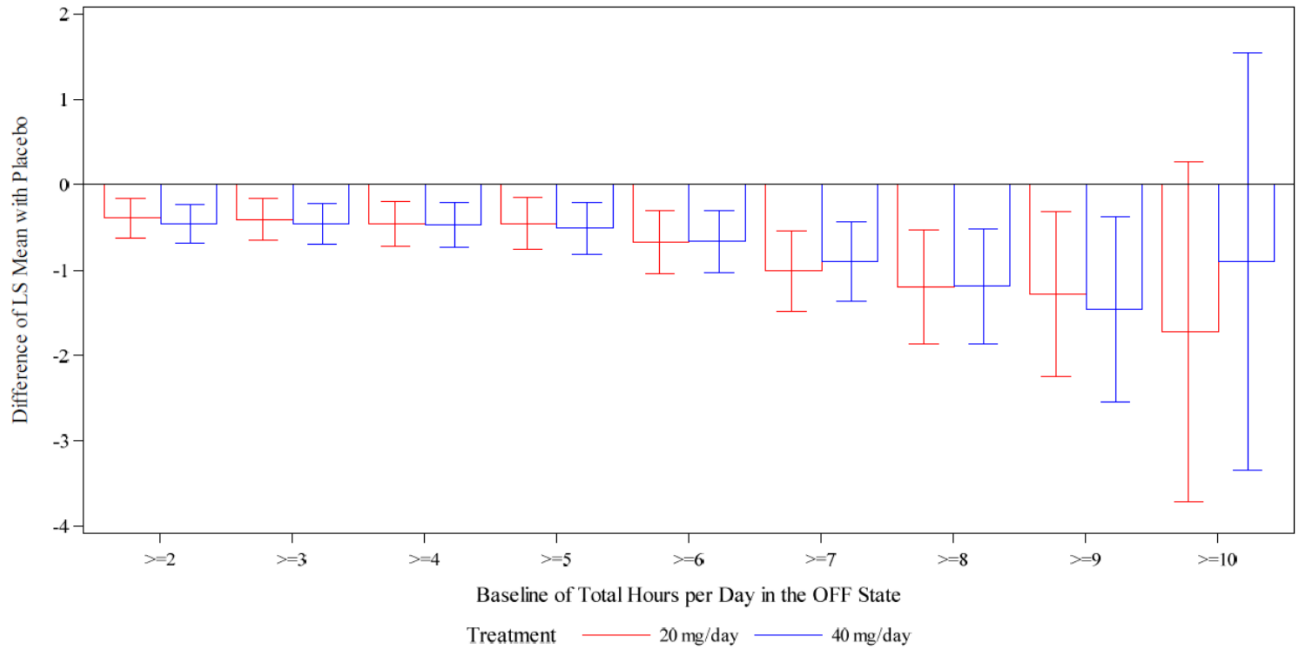
Expert physicians in PD were asked for their views regarding the survey results. The experts have endorsed the results from the patient surveys regarding the clinical relevance and veracity of the 20- to 30-minute treatment benefit.

European Parkinson's Disease Association (EPDA) has also reviewed the data presented as results of the Patient Perspective on "Off-Time" patient survey carried out by Kyowa Kirin. Overall, EPDA conclude that the results of this survey, which included a sample of more than 100 people with Parkinson's across five European countries, clearly demonstrate the importance of the management of "off-time" for people living with the condition.

Further Evidence of Clinical Benefit: Analysis of Effect by Baseline OFF Time

The applicant has presented a new subgroup analysis showing the LS Mean Difference from Placebo in the Change from Baseline to Week 12 Hours/day in the OFF State for istradefylline across baseline OFF time groups defined as cumulative 1-hour increments (Figure 48).

Figure 48: LS Mean Difference from Placebo in the Change from Baseline to Week 12 Hours/day in the OFF State [95%CI] for Istradefylline (20 mg/day and 40 mg/day) According to Baseline Hours of OFF Time - (OC, ITT, MMRM, Pool E1)



Source: Module 5, Re-exam Figure 5.1.

The applicant concluded that patients with the highest unmet medical need in terms of OFF time, i.e., those who are least well controlled on their current medications experience the highest degree of benefit with istradefylline on top of existing treatments for PD in a consistent manner across the istradefylline 20 mg/day and 40 mg/day dose cohorts when compared to placebo.

CHMP discussion on ground for re-examination #4

On 25 July 2021, the CHMP concluded that the efficacy of Nouryant has not been demonstrated considering the inconsistency of the results across the development program. Three grounds for refusal were adopted, highlighting specific methodological limitations of the efficacy dossier that prevented a conclusion on the validity of the estimate of the treatment effect of Nouryant in patients with PD. As a response to these grounds, the applicant submitted the grounds for re-examination including an additional ground discussing clinical relevance. The assessment and the discussion of the clinical relevance should be only applicable to reliable estimates of treatment effect. Therefore, the conclusion on the assessment on clinical relevance should be considered in relation with the conclusions of the prior three grounds on the methodological limitations.

In their response, the applicant has re-iterated previously presented data including the surveys and provided a new post-hoc subgroup analysis based on baseline OFF time in the pooled data population.

Istradefylline Treatment Effect

The results from the random-effects meta-analyses were already presented as part of the evaluation procedure prior the CHMP opinion on 22 July 2021. The mean improvement per day in OFF time over placebo is estimated to be 27 minutes for istradefylline 20mg and 28 minutes for istradefylline 40mg, but it could be as low as 7 minutes for istradefylline 20mg or 9 minutes for istradefylline 40mg. Although nominally significant, the modest effect sizes and the unadjusted wide nominal confidence intervals provide weak evidence for a clinically relevant effect.

Patient Surveys, Clinical Expert Statements and European PD Association Letter of Endorsement

Patient Surveys

These surveys (2 commissioned by the applicant) were already presented as part of responses during the evaluation of the procedure prior to the CHMP opinion on the 22 July 2021 and further discussed during the oral explanation the before the CHMP during meeting on 23rd June 2021.

- The quantitative survey of European PD patients was conducted and analysed by Adelphi Real World Research. It was conducted in several European countries (Spain, France, Germany, UK) in patients taking PD medications and experiencing OFF time. The first 14 questions of the survey were related to patient characteristics. Questions 17 and 19-22 are considered by the applicant as relevant to assess patient's perceptions of OFF and ON time. Although the responses were categorised as "no difference", "little difference", "moderate difference", "quite a difference" and "extreme difference", they are presented in this report into only three categories "no difference", "little difference" and "moderate-extreme difference", as the last one includes "moderate difference", "quite a difference" and "extreme difference". 66% of patients completed the survey themselves, and 34% completed it with carer support. Most patients were male (72%); most of them (78%) were taking 2 or more PD medications; around 90% had being diagnosed at least 3 years before completing the survey; 51% had OFF time every day; 60% of patients experienced a total OFF time of one hour or less while only 16% experienced an OFF time of 3 or more hours.

- Interviews with PD patients: This second survey was conducted in France, Germany, Italy, Spain and the UK by ThinkGen, in PD patients with ongoing experience of OFF episodes or OFF time that impacted their daily routines and activities and their caregivers. Thirty-six semi-structured interviews were conducted in April 2021. 53% of participants were male with a mean age of 62.4 years of age; 59% of them had 1-2 episodes per day of OFF time. Patients were selected by patient groups and healthcare providers from a group of individuals who have previously expressed interest in participating in market research.
- Parkinson's disease and Movement disorder Alliance Survey: The survey was conducted in February 2021 via an electronic newsletter. There were 408 respondents who either have PD or are a loved one of someone with PD and aimed to understand how a reduction in OFF time could improve quality of life. 284 respondents were identified as patients. Only patient perceptions on the value of OFF time of 30 minutes and of 1 hour are presented.

Based on the results from the EU surveys, the applicant concluded that a decrease of 20 or 30 minutes in OFF time or a similar increase in ON time without troublesome dyskinesias (Adelphi survey only) could be considered clinically relevant. However, perception of benefit may be affected by the amount of time a patient spends in the OFF state. In fact, a lower proportion of patients felt a decrease of 20 minutes in OFF time would be moderately-extremely beneficial (68%) in the group of > 2 hours OFF time compared to experiencing ≤ 2 hours OFF (82%) as displayed by the results of the Adelphi survey. In this regard, the study population included in the EU surveys was substantially different in terms of average daily OFF state from the one included in the istradefylline trials. In the pivotal studies, the mean time per day spent in the OFF state was 6.22 hours ranging from 5.4 hours (mean) in Study 6002-014 to 6.7 hours (mean) 6002-US-018. In the Adelphi Real World Research survey, only 5% (n=5 patients) experienced a total OFF time of six or more hours per day while 60% experienced a total OFF time of one hour or less during a 24 hour period. Similarly, only 6% (n=2 patients) of the patients experienced a total OFF time of five or more hours during a 24 hour period whereas 59% experienced a total OFF time of one-two hours per day in the ThinkGen survey. This aspect has been acknowledged by the applicant who noted that it was intended to recruit a broader PD population in the surveys so it represents the broad population of PD patients who could become eligible for istradefylline treatment in the event of a Marketing Authorization Approval in Europe. However, the position of the applicant that the studied population included a significant overlap with the istradefylline pivotal study population cannot be agreed. Due to this it is concluded that survey results cannot readily be extrapolated to the populations in the pivotal studies.

The following statement in the ground for re-examination is noted: Please note that the applicant has undertaken a follow up Adelphi survey exclusively with patients experiencing ≥2 hours OFF time on any given day. Preliminary results (from 58 participants to date) are highly consistent with the previous survey. These data are available upon request." However, it should be considered that content of the grounds should be based only on scientific data available at the time of the initial Opinion. As such, the follow up data on subjects with ≥2 hours OFF cannot be considered as part of this re-examination.

As regards the US survey, no information is provided about the total OFF time of the included patients. On note, respondents included in this survey were additionally asked to value the impact of 1 hour less of OFF time, which is aligned with MCID of 1 hour OFF time proposed by Hauser (2014) as a threshold value for clinical relevance.

Additionally, the applicant states that they made efforts to avoid bias in the questionnaire but questions 19 to 22, asking specifically about the advantage of getting 20 to 30-minute decrease in OFF time, include sentences like "…with very few additional side effects" or "…would this make a positive difference to you?". The response bias (acquiescence bias) cannot be ruled out here as the respondents can be affected by these statements and tend to agree with them. Moreover, Questions Q19-Q22 as formulated

in the Adelphi Survey (see above), linked the improvement in ON/OFF time to a new medication with few additional side effects. While it can be agreed that the safety profile could be considered manageable with the appropriate risk minimization measures, several adverse-drug reactions were identified for Nouryant. Additionally, cases of cardiac SAEs were presented by the applicant which were considered as possibly related to istradefylline and the applicant was requested to commit monitoring of any signal on cardiac events suggestive of potential causal relationship with istradefylline treatment in the post-marketing setting. Finally, the uncertainties about the long-term safety profile in particular given the absence of evidence from the dose findings studies supporting the proposed dose of 40mg cannot be ignored. In addition, patients were selectively approached by their respective physicians and patient groups as possible participants that they expected to be eligible. This would result in a too optimistic picture of the attitude that is measured and have a relevant impact on the validity of the survey. Also the fact that it is a self-completed survey and data provided by the patients were not checked with medical records further limits the validity and quality of this survey.

Clinical Expert Statements

The applicant has submitted testimonials from five neurologists supporting the benefit of a decrease in OFF time of around 30 minutes and an increase in ON time without troublesome dyskinesia of 20 to 30 minutes. These letters were already included as during the evaluation prior of CHMP opinion on 22 July 2021 and were considered supportive of the way a clinically relevant effect could be defined in the specific population.

European PD Association Letter of Endorsement

Overall EPDA´s conclusion on the importance of the management of "off-time" for people living with the condition is acknowledged. In fact, the CHMP Guideline on clinical investigation of medicinal products in the treatment of Parkinson's disease (EMA/CHMP/330418/2012 rev. 2) already includes "OFF"-time or "ON"-time as possible main efficacy variables. The importance of reducing "OFF" time as a therapeutic goal for symptomatic therapies is not at discussion at this particular stage. What is questioned and represents a key point in the assessment of this additional Ground for Refusal is the effect size of this reduction.

Further Evidence of Clinical Benefit: Analysis of Effect by Baseline OFF Time

The new post-hoc subgroup analysis presented by the applicant suggests a direct association between baseline OFF time and magnitude of effect for patients with baseline OFF time $\geq$ 6 hours per day. Below this threshold, the exploratory analysis does not support a linear trend. This association was already presented previously by the applicant using median OFF time per day per region as a cut-off for dichotomization (subgroup analysis report) (Table 143).

Table 143: Baseline Total Hours/Day in the OFF State Subgroup by Regional Median. Difference in LS Means (95%CI) vs Placebo, Change from Baseline at Week 12 Total Hours per Day in the OFF State, MMRM: ITT Analysis Set

Population	Baseline Total Hours/Day in the OFF state (hours) (N Istra 20,40 mg /placebo)	Difference in LS mean (95% CI), 20 mg istradefylline	Difference in LS mean (95% CI), 40 mg istradefylline
Europe	<5.5 hours (N=42, 61/N=51)	-0.23 (-0.97, 0.51)	-0.25 (-0.93, 0.44)
	≥5.5 hours (N=36, 51/N=60)	-0.73 (-1.93, 0.47)	-0.53 (-1.58, 0.52)
North American	< 6.3 hours (N=229, 149/N=224)	0.01 (-0.40, 0.41)	-0.30 (-0.76, 0.16)
	≥6.3 hours (N=240, 183/N=237)	-0.65 (-1.15, -0.15)	-0.45 (-1.01, 0.10)
Japan	<6 hours (N=107, 115/N=111)	-0.73 (-1.25, -0.21)	-0.29 (-0.79, 0.22)
	≥6 hours (N=109, 112/N=106)	-0.92 (-1.59, -0.25)	-1.28 (-1.94, -0.61)

Results from the new subgroup analysis are difficult to interpret in terms of causal effects. Firstly, this is a post-hoc analysis and no adjustment for multiplicity was performed. In this regard, it can only be considered as an exploratory analysis. Secondly, the figure suggests that an association may exit for those with baseline OFF time ≥ 6 hours per day. However, the wide 95%CI for the corresponding estimates indicate a high degree of uncertainty of effect size. Finally, it is difficult to conclude on the type of population that patients with the highest level of baseline OFF time are representing in the pooled data analysis. These patients may represent a population of advanced PD who in spite of the best therapeutic efforts may still have a therapeutic need for managing OFF time, but could also represent a population with suboptimal therapeutic control of OFF time at study entry. During the evaluation prior to CHMP opinion on 22 July 2021, the applicant explained that patients were receiving optimal treatment prior to trial participation on the basis of levodopa dose and use of adjunctive therapies at entry, and the fact that patients were recruited from centres specialising in movement disorders. This argumentation can be followed, but it should be noted that therapeutic management has evolved over time and the istradefylline drug development program extended from 2002-2016. For example, the initiation of levodopa in early patients should not be delayed based on a potential deleterious effect on PD progression (oxidative stress) and the increased risk of motor complications, as new evidence suggest that there is no evidence for toxicity (Verschuur CVM et al NEJM 2019) and it could be a modest benefit (PD Med Collaborative Group Lancet 2014). The use of non-pharmacological approaches (DBS) are becoming more accessible and recommended for treatment of motor complications. It is expected that the direction goes to a use in an early stage (SH Fox et al. Mov Disord 2018). As per regards of treatment of wearing off, it is recommended that DA should be used at maximum tolerated dose (Zvezdan Pirtošek et al. Parkinson's Disease, vol. 2020). In fact, the inclusion criteria for Study 6002-014 (2013-2016) required patients currently taking levodopa combination (carbidopa/levodopa or benserazide/levodopa) therapy with a total daily levodopa dosage of at least 400 mg plus a recommended, clinically effective dose of at least one adjunctive medication approved to treat Parkinson's disease. Even if the other trials allowed the use of other symptomatic drugs for motor complications, the requirement of using at least one adjunctive drug at a clinically effective dose was only included for Study 6002-014. In this regard, the absence of positive results in the most recent study (Study 6002-014; 2013-2016) in 'maximally and optimally treated' patients showing the lowest baseline OFF time across the trials in spite of the largest mean time since onset of motor complications is a concern that cannot be ignored.

Overall conclusion on the fourth ground of re-examination on Clinical Relevance

To support the relevance of the effect the applicant has provided some evidence from patient surveys suggesting that a decrease of 20-30 minutes in OFF time or similar increase in ON time without troublesome dyskinesia, is beneficial. However, such surveys are not free of potential bias. In addition, the baseline time in OFF state is key to understand the relevance of the gained time (it is not the same to reduce 30 minutes in OFF time in 1 hour that 30 minutes in 6 hours). In the quantitative survey, 60% of respondents had a mean daily OFF duration of $\leq$ 1 hour making such results hardly generalizable to the patients included in the pivotal trials for whom the mean number of hours spent in OFF state ranged from 5.36 to 6.7 hours/day.

Considering all of the above it should be reiterated that the assessment of data and any discussion on the clinical relevance of a possible effect in this population, should only be possible in case a reliable treatment effect has been concluded on. Even so, the data and argumentations provided by the applicant as part of the re-examination procedure were reviewed and the assessment conclusions are that they cannot be considered sufficient evidence to support the clinical relevance of the effect sizes, seen in the pooled pivotal studies.

5.1.5. SAG-N meeting on istradefylline

The experts are invited to provide their views on the CHMP grounds for refusal, taking into account the company's response:

The applicant submitted eight pivotal clinical studies in support of this application. While four of the eight pivotal trials submitted met their primary endpoint of a reduction in the % change from baseline to endpoint in time spent OFF (studies US-005, US-013) or change from baseline to endpoint in total hours spent OFF (Japanese studies 0608 and 009), the other trials did not.
Therefore, the efficacy of the product is not demonstrated considering the inconsistency of the results across the development program. More specifically:

- **Study US-006 was a formally failed study as per the pre-specified primary ANOVA analysis model. The primary analysis model was changed from ANOVA to ANCOVA after finalising the SAP and even with changing the analysis model the primary endpoint result was marginal, achieving a nominal p-value of p=0.049. Considering the late change in the statistical analysis, this study is not considered to have demonstrated a statistically significant treatment effect.**
- **The inconsistency of study results including the different responses observed in the studies in different regions and the difference in response over time, are not resolved by post-hoc pooling the individual trials intended to rescue their unconvincing results. In addition, the estimated modest effect size, and the unadjusted nominal confidence interval that is in close proximity to no effect are not considered sufficient to have demonstrated a treatment effect in a post-hoc pooled analysis. Moreover the observation of no clear pattern of a dose response with increasing doses of istradefylline, leads to considerable uncertainty on the effect of the treatment.**

- **The fact that Studies 007 and 014 (that enrolled the European population) both clearly failed remains an unresolved issue. In particular, as the most recent study, Study 6002-014 in 'maximally and optimally treated' patients did not show positive results – despite being the largest pivotal trial relative to size of each treatment group and despite being planned, taking into account the results of previously conducted istradefylline's trials.**

The experts endorsed by consensus the grounds of refusal for a negative B/R for istradefylline as presented by the CHMP, although two of them suggested that a signal of efficacy of the medication could be retrieved from the 4 positive trials.

It was noted indeed that the explanations provided by the applicant about the negative results generated by some of the trials and their heterogeneity, were not convincing. In particular were of concern the negative results observed in the studies including a European population. In this relation, it was mentioned that in previous studies the Japanese population differed as for posology needed to handle PD symptoms and risk for adverse events. This heterogeneity should be considered evaluating the positive results obtained in the Japanese populations, not confirmed in the studies including a European population.

One expert noted that the results of the studies could be compatible with a marginal effect size and a discussion about its clinical relevance pointed out that it could not be considered as clinically relevant. An expert questioned validity of the survey submitted by the applicant to the patients and highlighted that the 20 minutes average benefit is below what it is considered as clinically relevant. This notion was confirmed by the patient representative who explicitly expressed that taking this medical product could imply additional adverse events for only 20 minutes of benefit in OFF time. Experts also mentioned that even the upper limit of 95%CI around pooled estimate for reduction in absolute hours OFF time was below 1 hour.

Finally, in general *post-hoc* analysis are considered by the experts appropriate only as hypothesis generating approach but not valid for confirmatory/conclusive results. It was noted that for US-006, the primary model was changed from ANOVA to ANCOVA after concluding statistical analysis plan and therefore, ANCOVA-results were considered as a post-hoc and data driven analyses.

In addition to providing their views on the CHMP grounds for refusal, the experts are invited to provide input on the following questions:

Methodological questions:

1. With reference to Study US-006, what is your view on the acceptability of the post-hoc change to an ANCOVA after unblinding as well as the validity of the predefined ANOVA in the randomised setting?

The experts believe ANCOVA is the optimal method for this analysis. However the change from ANOVA - the method predefined in the statistical analysis plan - was done after finalizing the statistical analysis plan. Therefore, the experts agreed that the results obtained with ANOVA are those to be considered in this evaluation whereas the results from ANCOVA should be considered only as data driven.

2. The development program of 8 studies included 4 positive studies and 4 trials that did not demonstrate efficacy. Please comment on the following:

a) What is your view on the heterogeneity of the trial populations and, if any, to what extent may this have impacted the results?

Experts noted that there may be sources of heterogeneity both internal (within each study) and external (among studies). In this regard, factors highlighted by the experts for this specific application were ethnicity, gender (higher female proportion in Japanese studies), BMI (lower BMI in Japanese studies), disease severity, background therapy. In particular as heterogeneity source it was noted the use of background therapies (evolved over the years) and differences in genetic polymorphisms of ADORA2A gene receptors between European and Japanese populations. It was pointed out that in any case, heterogeneity should have been considered in advance in the design of the trials (increased variability -> decreased statistical power).

b) Can the meta-analysis as performed be regarded as robust clinical evidence of efficacy and safety of istradefylline in patients with Parkinson's disease, and how do you consider the level

of evidence provided by the meta-analysis as compared to the standard requirements? Please outline your arguments.

The experts do not consider that the performed meta-analysis could be regarded as a robust clinical evidence of efficacy and safety as compared with the standard requirements (individual pivotal trials). Additionally, it was emphasized that the analyses were done as a *post-hoc* approach. In addition, these were not properly performed (not pre-defined, multiple endpoints, lack of multiplicity control). Finally, effect size was found to be marginal.

c) What is the impact on the interpretation of the results of other aspects inherent to the post-hoc meta-analysis, e.g. the lack of multiplicity control for the two doses?

The lack of multiplicity control for the two doses is considered one of the key study limitations.

Strictly speaking about the two doses, the Bonferroni-corrected p-values would be still below 0.025 (e.g. 0.05/2). However, it should be noted that on top of the two doses, there were many other choices made in this post-hoc analysis: which studies to pool, which analysis method to use, and different endpoints. All such choices were not taken into account into the multiplicity control.

d) Given the proximity of the nominal confidence interval to 0, what is your opinion on whether it is secured that 'no effect' can be excluded in light of the post-hoc analysis?

Overall, experts believe that the effect size-based on 95%CI not including "zero"- could be considered as different from zero but the probability that this is true is not granted for the above described reasons. In addition it was reiterated that was a post-hoc analysis. Lack of multiplicity control should be considered as an additional limitation to the robustness of these results. Moreover, the effect size cannot be considered as clinically relevant.

Finally, considering the large number of patients who participated in the different studies included in the meta-analyses, it is expected that 95%CI could not include 0.

Remarkably, based on the available evidence and the presence of other available alternatives, a patient representative in the Group mentioned that taking this additional drug would be a concern unless a new trial conducted in a European population shows efficacy. This drug could be taken as a participant of an investigational trial.

Clinical questions:

3. What is your view on the representativeness of the trial populations for today's European patients considering the heterogeneity of the study populations, in terms of the inclusion/exclusion criteria (in particular the need to be effectively treated with Levodopa (plus an adjunctive medication)), and the regional and temporal spread.

The number of patients included in the 8 pivotal trials could be considered as representative of PD population who could benefit from this drug. Experts noted that the population included in some of the trials had a very severe disease form and this could have negatively impacted the results. Again, the heterogeneity (Japanese vs. European) was noted and lack of significant results in the studies including European population was again highlighted. It was also mentioned that type of drugs and the way they are used for managing PD has clearly evolved over the last 20 years, a fact which impacts the interpretability of the results of studies conducted many years ago.

4. If the methodological constraints to conclude on efficacy for the individual studies and the meta-analysis would be set aside,

a) **Do you consider that the observed effects on the primary and secondary endpoints are sufficiently robust and clinically meaningful taking into account that the trends for beneficial effects were most prominent in the Japanese studies (studies 6002-0608 and 6002-009) but were not confirmed for the European population in studies 6002-014 and 6002-007?**

Experts unanimously agreed that effects on the primary and secondary endpoints are not robust and not clinically relevant enough, in particular, considering heterogeneity and the lack of effect in studies including European population.

b) **In this respect, what is your view on the clinical relevance of the responder analysis results in study 6002-014 (which was specifically designed to show that istradefylline treatment on top of optimized PD background therapy would result in added benefit for the patients) where the percentage of responders (reduction in OFF time ≥ 1 hour AND increase in ON time without troublesome dyskinesia ≥ 1 hour) was higher in placebo (39%) that in the two istradefylline groups (20 mg: 35%, 40 mg: 34%)?**

Experts considered that negative results from Study 6002-0014 are of the highest importance to drive the position about the clinical relevance of istradefylline because it included a maximally treated population as per inclusion/exclusion criteria, European patients and it was the study with the largest sample size. In this regard, the add-on effect of istradefylline suggested by other studies is not confirmed in this responder analysis. It was actually noted that the results from this study do not exclude a negative effect (placebo might still be better than istradefylline).

5.1.6. Oral explanation

The applicant presented at an oral explanation the grounds for re-examination.

During the presentation, the applicant claimed that a positive trend was seen in 7 out of the 8 pivotal trials supporting in ther views, an overall positive istradefylline treatment effect from individual studies. As for study 6002-US-006, the applicant stated that ANOVA remains the primary analysis as reported in the CSR and that regardless of the method (ANOVA or ANCOVA), there was a clear positive OFF-time treatment.

As per the pooled analysis, the applicant presented the results and claimed that they do comply with prerequisites in the CHMP Points to Consider on application with 1. meta-analysis; 2. one pivotal study (CPMP/EWP/2330/99).

Heterogeneity and lack of responses in EU studies was also discussed. In this regard, the applicant claimed that there were not fundamental differences between the Japanese and EU populations, in particular no differences in genetic polimorphims and standard of care and reiterated that the results from the Japanese studies are not inconsistent with the results across the programme as a whole. Additionally, they presented subgroup analyses (Europe vs. Overall) for OFF state and for responder analyses to support efficacy in European population.

Finally, the applicant presented the differences from placebo in the change from baseline to week 12 Hours/day in the OFF State by cumulative subgroups by baseline OFF-time to support that the benefit is higher for the subgroups with the highest baseline OFF-time.

During the discussion, the CHMP reiterated that positive trends in the primary variable were not seen in two comparisons that estimated literally no effect as per the point estimate as well as the one that estimated a negative effect and therebyl this requirement in CPMP/EWP/2330/99 is not meet. As per the criterion of "Pooled 95% confidence interval well away from zero", considering that the unadjusted lower

limit of the confidence intervals ended at 7 resp. 9 minutes, the CHMP again reiterated that this criterion is not considered met.

The CHMP reinforced that in spite of subgroup analyses, it remains a fact that pivotal studies including European population (6002-EU-007 and 6002-014) did not meet the primary endpoint.

Finally, the applicant was invited to comment which PD population represents the subgroup patients with highest level of OFF time at baseline. It was noted that level of off time at baseline could be in relation with severity but also with the background therapy (optimal vs. suboptimal). This group of patients could present a PD population without an optimal treatment (levodopa + adjunctive therapies). The applicant replayed that for study 6002-014 the pattern was the same. However, it remains a fact that study 6002-14 included a population with optimal PD treatment (as per inclusion criteria) who in spite of showing largest the time since onset of motor complications showed the lowest mean of hours in OFF at baseline (5.4 SD(2.01)).

5.1.7. Overall conclusion on grounds for re-examination

The CHMP assessed all the detailed grounds for re-examination and argumentations presented by the applicant and considered the views of the Scientific Advisory Group.

The original application contains eight pivotal clinical studies with four positive studies (studies 6002-US-005, 6002-US-013, 6002-0608 and 6002-009) and four negative studies (6002-US-006, 6002-US-018, 6002-EU-007, 6002-014), so there is inconsistency in the conclusions drawn on a treatment effect. Additionally, no clear dose-response could be established across the 8 pivotal trials. For study 6002-US-006, the applicant argues that ANCOVA would have been the most appropriate analysis for a continuos change from baseline endpoint. However, the fact remains that ANOVA was prespecified and the switch to ANCOVA was made post-hoc, an argument agreed by experts during the SAG-N.

Given the inconsistency of the individual study results the applicant undertook a pooled analysis of the 8 pivotal studies. The applicant argues that this analysis of the eight studies is sufficient based on the prerequisites in the CHMP Points to Consider on application with 1. meta-analysis; 2. one pivotal study (CPMP/EWP/2330/99). However, the pooled analysis had not been predefined, but was rather performed as a rescue measure following the observation that the individual trials, which were planned to stand on their own, showed only borderline or no effect. Next to various methodological concerns, the estimated effect size with the random effects meta-analysis was modest and while the 95%-CIs excluded '0', given the proximity of the CI-boundaries to 'no effect', a clinically relevant treatment effect cannot be confirmed. This precludes the use of the meta-analysis as pivotal evidence for the MAA in a situation where also the results from eight individual studies did not allow to conclude that efficacy was established due to inconsistent effects. This argument was agreed by the experts in the Scientific Advisory Group Neurology (SAG-N) who highlighted that the pooled analysis has methodological weaknesses (not pre-defined, lack of multiplicity control) and even putting methodological limitations aside, the estimated magnitude of the effect was found to be marginal, not clinically relevant.

While the applicant considers Study 6002-014 showed signs of effectiveness, this is not agreed by the CHMP. This trial was performed to show an effect in maximally and optimally treated patients and also included European patients and is considered one of the most important among the pivotal studies. The results of the primary and secondary endpoints were negative or inconclusive, and post-hoc analyses cannot solve this problem.

Both studies including EU population (6002-14 and 6002-EU-007) were negative. In order to address CHMP concerns around the efficacy of istradefylline in the European population, the applicant conducted a number of post-hoc and regional subgroup analyses. However, post-hoc analyses cannot be accepted as pivotal evidence upon inconsistent or negative results in the primary and secondary analyses.

Furthermore, the post-hoc analyses results are also not convincing from a clinical point of view. Finally, the region analysis only increases the concerns regarding a potential effect of istradefylline in the European population. Experts in the SAG-N unanimously agreed that the absence of positive results in the studies including EU populations is a concern. Finally, during the SAG-N experts were asked about the potential sources of heterogeneity of the trial populations. Experts highlighted the differences in use of background therapies that has evolved over the years. In this regard, studies 6002-US-005 and 6002-US-013 were conducted between 2002-2003, over 15 years ago, making their external validity and generalisability to current treatment settings questionable. The other two positive studies-Studies 6002-0608 and 6002-009- were conducted in Janapense populations. Althogether, the claim made by the applicant that results in Nouryant can be extrapolated to EU population cannot be accepted.

Finally, the applicant submitted an additional ground of re-examination that the effect size observed in Nouryant drug development plan can be considered clinically relevant. To support this claim, the applicant reiterated the results from patient surveys suggesting that a decrease of 20-30 minutes in OFF time or similar increase in ON time without troublesome dyskinesia, is beneficial. However, such surveys are not free of potential bias. In addition, the baseline time in OFF state is key to understand the relevance of the gained time (it is not the same to reduce 30 minutes in OFF time in 1 hour that 30 minutes in 6 hours). In the quantitative survey, 60% of respondents had a mean daily OFF duration of $\leqslant$ 1 hour making such results hardly generalizable to the patients included in the pivotal trials for whom the mean number of hours spent in OFF state ranged from 5.36 to 6.7 hours/day. At least 1 hour of reduction in OFF time was proposed as the MCID by Hauser, 2014 and was confirmed by SAG-N experts who unanimously agreed that effects on the primary and secondary endpoints observed for Nouryant are not clinically relevant.

6. Benefit-risk balance following re-examination

6.1. Therapeutic Context

6.1.1. Disease or condition

Parkinson's Disease is a progressive, debilitating movement disorder characterised by bradykinesia, rigidity, and resting tremor that affects 1.2 million people in Europe (European Brain Council, 2019). Most motor symptoms of PD result from the progressive degeneration of the dopaminergic neurons in the substantia nigra pars compacta. As degeneration occurs, striatal concentrations of dopamine decrease, leading to reduced stimulation of dopamine receptors in the striatum. Dopamine activates the 'direct pathway' from the striatum of the basal ganglia via D1 receptors and suppresses the 'indirect pathway' from the striatum via D2 receptors. Thus, the reduced striatal dopamine that occurs in PD results in decreased activity of the direct pathway and increased activity of the indirect pathway. Increased excitability of the indirect pathway can be mediated by adenosine A_{2A} receptors in the striatum. The imbalance of activity between the direct and indirect pathways in PD is thought to result in the hallmark symptoms of bradykinesia, rigidity, tremor, and loss of postural reflexes.

6.1.2. Available therapies and unmet medical need

Levodopa remains the "gold standard" for therapy for PD that increases dopamine concentrations in the brain because levodopa can cross the blood-brain barrier and be transformed into dopamine. In the early stages of PD, patients usually experience substantial symptom relief from levodopa. Despite its initial efficacy, levodopa's therapeutic window narrows over time (Jankovic, 2005). That is, the duration of benefit from a dose of levodopa ("ON time") becomes shorter and PD symptoms return before the next

scheduled dose ("wearing-off"). There are periods of time when, despite measurable plasma concentrations, levodopa does not control PD symptoms ("OFF time") and these periods become increasingly longer as PD progresses.

Efficacy of available treatments varies from patient-to-patient and over time. There are many different types of dopaminergic therapies that have been developed to try to treat OFF time (e.g., dopamine agonists), COMT inhibitors, MAO-B inhibitors); however, treatment is complicated by an increased risk of dyskinesia and a variety of side effects, including impulse control disorders and sleep disturbances, which may limit benefits or preclude continued use of these medications. Thus, despite available medical therapies, patients continue to suffer potentially disabling OFF episodes. Consequently, there is a continuing need for additional agents that are effective for the levodopa-treated patient (producing less OFF time and better symptom control) without intolerable side effects.

The clinical heterogeneity of PD signs and symptoms and in the individual levodopa requirements reinforces the need for additional adjunctive treatment options (Lewis, 2005).

Istradefylline (also known as KW-6002) is an adenosine A_{2A} receptor antagonist, which has a xanthine derivative structure and which competitively inhibits adenosine binding to the A_{2A} receptor. As an adenosine A_{2A} receptor it would represent an add-on treatment with a new mechanism of action for PD patients.

The target indication for istradefylline was as an adjunctive treatment to levodopa-based regimens in adult patients with PD experiencing "OFF" time.

6.1.3. Main clinical studies

The applicant has included 8 pivotal Phase 2b/3 randomized, double-blind, fixed-dose, placebo-controlled studies (6002-US-005, 6002-US-006, 6002-US-013, 6002-0608, 6002-009, 6002-US-018, 6002-EU-007, and 6002-014) and 2 open-label, long-term studies (6002-US-007, 6002-010) (Table 144). These studies in PD patients evaluated the safety and efficacy using 10, 20, 40, and 60 mg once daily doses of istradefylline. The applicant also included a retrospective meta-analysis of the 8 pivotal studies.

Table 144: Study design comparison

6002-US-005	6002-US-006	6002-US-013	6002-0608	6002-009	6002-US-018	6002-EU-007	6002-014
Design: Double-blind, randomized, placebo-controlled, parallel-group clinical study							
Duration: 12-week	12-week	12-week	12-week	12-week	12-week	16-week	12-week
Treatment Groups (randomization ratio):							
Istradefylline 40 mg/day or placebo (2:1 ratio)	Istradefylline 20 or 60 mg/day or placebo (2:2:1 ratio)	Istradefylline 20 mg/day or placebo (1:1 ratio)	Istradefylline 20 or 40 mg/day or placebo (1:1:1 ratio)	Istradefylline 20 or 40 mg/day or placebo (1:1:1 ratio)	Istradefylline 10, 20, or 40 mg/day or placebo (1:1:1:1 ratio)	Istradefylline 40 mg/day, or placebo, or entacapone (1:1:1 ratio)	Istradefylline 20 or 40 mg/day or placebo (1:1:1 ratio)
Subjects were: • At least 30 years of age (at least 20 years of age in Studies 6002-0608 and 6002-009); • Diagnosed with PD as determined by the UKPDS criteria; • Modified Hoehn and Yahr scale Stages 2 to 4 in the OFF state (Studies 6002-US-005, 6002-US-006, 6002-US-013, 6002-0608, 6002-009, 6002-US-018, and 6002-EU-007), and in the ON state (Study 6002-014); and • Had end-of-dose wearing-off with an average of at least 2 hours OFF time per day (Studies 6002-US-005, 6002-US-006, 6002-0608, 6002-009, and 6002-014) or 3 hours OFF time per day (Studies 6002-US-013, 6002-US-018, and 6002-EU-007) at study entry; subjects in 6002-014 also had levodopa-induced dyskinesia.							
Levodopa Requirements: • Receiving levodopa and a peripheral DOPA-decarboxylase inhibitor (carbidopa or benserazide) for at least 1 year. • Treated with levodopa for at least 1 year and have been on a stable regimen of levodopa for at least 4 weeks before randomization/baseline (as per protocol specification). – Studies 6002-US-005 and 6002-US-006: at least 4 doses/day, or at least 3 doses/day if 2 doses were slow-release formulations (no specific levodopa dose requirement). – Studies 6002-US-013, 6002-US-018, and 6002-EU-007 at least 3 doses/day (no specific levodopa dose requirement). – Studies 6002-0608 and 6002-009: at least 300 mg/day levodopa. – Study 6002-014 required subjects to be taking at least 400 mg/day levodopa plus at least 1 adjunctive dopaminergic medication approved to treat PD, and with documented levodopa-induced dyskinesia. • Decrease in the total daily dose of levodopa was permitted, if necessary (Investigator's discretion) due to levodopa-related AEs. • Change in either the frequency of levodopa dosing or the interval between levodopa doses was not allowed (Exception: Study 6002-EU-007 allowed levodopa dose adjustment during the initial 4 weeks).							

6002-US-005	6002-US-006	6002-US-013	6002-0608	6002-009	6002-US-018	6002-EU-007	6002-014
Other Parkinson's Medications were allowed (and were required in Study 6002-014). • Reduction in the dose of anti-parkinson medication was permitted to control dopaminergic-related AEs if a prior reduction in the levodopa dose was unsuccessful, or to alleviate AEs thought to be directly related to the agent being adjusted. • Prior anti-parkinson medication could not be increased and no new anti-parkinson medication could be added.							
Primary Endpoint (per protocol): Change from baseline in OFF time/day expressed as:							
% of awake time	% of awake time	% of awake time	Hours	Hours	% of awake time	% of awake time	Hours
Primary Statistical Methodology: Analysis Set[a]/Analysis Method (per protocol and SAP)							
ITT/ANOVA, ANCOVA[b] 12 week-LOCF, OC	ITT/ANOVA, ANCOVA[b] 12 week-LOCF, OC	ITT/ANCOVA 12 week-LOCF, OC	FAS/ANCOVA 12 week-LOCF, OC	FAS/ANCOVA 12 week-LOCF, OC	ITT/ANCOVA 12 week-LOCF, OC	ITT/ANCOVA 16 week-LOCF, OC	ITT/MMRM 12 week, OC

a the ITT and FAS analyses were essentially the same.
b The SAP prespecified ANOVA and the protocol prespecified ANCOVA
AE= Adverse event; ANCOVA=analysis of covariance; ANOVA=Analysis of variance; FAS=Full analysis set; ITT=intent-to-treat; LOCF=Last observation carried forward; MMRM=Mixed-effects model repeated measures; OC=Observed Case, PD=Parkinson Disease; SAP=Statistical analysis plan; UKPDS=United Kingdom Parkinson´s Disease Society.

6.2. Favourable effects

Four of the eight pivotal trials, met their primary endpoint showing a favourable reduction in the % change from baseline to endpoint in time spent OFF (studies US-005, US-013) or change from baseline to endpoint in total hours spent OFF (Japanese studies 6002-0608 and 6002-009). For reducing daily OFF time, a random-effects meta-analysis showed an overall treatment effect equates to a mean 27-28 minutes improvement per day in OFF time over placebo in the overall pivotal study population, a reduction that according to the patient´s surveys and experts opinion could be considered clinically relevant. According to the applicant, patients with highest baseline OFF time could experience a benefit larger than the average 27-28 minutes of OFF reduction.

As per the most relevant clinically secondary endpoint, i.e. change in ON time without troublesome dyskinesia, two trials showed nominally significant differences in favour of istradefylline (Study 6002-US-005, Study 6002-009).

6.3. Uncertainties and limitations about favourable effects

The original application contains eight pivotal clinical studies, with four positive studies and four negative studies, so there is inconsistency in effect.

Study 6002-US-018 did not meet its primary endpoint and in fact, this study demonstrated numerical worsening for istradefylline 10mg and 20mg compared to placebo with only a very minor numerical improvement for istradefylline 40mg compared to placebo that was not statistically significant nor clinically meaningful with a reduction in OFF time compared to placebo of 0.03 hours for istradefylline 40mg. For the secondary endpoint, ON time without troublesome dyskinesia, in study 6002-US-018 for all istradefylline doses 10, 20 and 40mg there was a numerical decrease found which could be considered clinically unfavourable. None of the secondary endpoints were controlled for multiplicity thus this was only considered descriptive.

Study 6002-EU-007 did not meet its primary endpoint Istradefylline 40mg failed to show any benefit over placebo in the only pivotal study conducted with an additional active comparator (entacapone 200mg daily). In fact, there was no difference between Istradefylline 40mg in percentage reduction of OFF from baseline at endpoint (-4.53% for placebo and -5.14% for Istradefylline 40mg) and placebo. The reduction from baseline for entacapone was somewhat greater at -7.82%. The difference versus placebo was however also not statistically significant.

While the applicant considers Study 6002-014 showed signs of effectiveness, this is not agreed. This trial was performed to show an effect in maximally and optimally treated patients and also included European patients and is considered one of the most important among the pivotal studies. The results of the primary and secondary endpoints were negative or inconclusive, and post-hoc analyses cannot solve this problem.

For study 6002-US-006, the applicant argues that ANCOVA would have been the most appropriate analysis for a continues change from baseline endpoint. However, the fact remains that ANOVA was prespecified and the switch to ANCOVA was made post-hoc. During the SAG-N, the experts agreed that the results obtained with ANOVA are those to be considered for the primary endpoint.

The two positive US studies (US-005 and US-013) were conducted between 2002-2003, over 15 years ago, making their external validity and generalisability to current treatment settings questionable.

Given the inconsistency of the results and conclusions from the individual studies that were planned to stand on their own the applicant undertook a retrospectively defined pooled analysis of the 8 pivotal studies, and argues that this meta-analysis is sufficient based on the prerequisites in the CHMP Points to Consider document on application with a meta-analysis (CPMP/EWP/2330/99). The Points to Consider states that "*a meta-analysis cannot be used to reconcile the conflicting results of one positive and one inconclusive study*", which could be extended to this case with four positive and four inconclusive studies and only three of them showing numerical improvement. While, there is some room opened to consider a meta-analysis as an exceptional case, such will always be problematic and it is also explained therein that the necessity to resort to a meta-analysis to reach an acceptable level of significance may question the clinical relevance. Specifically in this application, several of the prerequisites (inconclusive studies with positive trends, well away from zero) specified in the points to consider are considered as not fully met. Furthermore, (modest) heterogeneity was observed.

The interpretation of the meta-analysis is hampered by the fact that it was performed retrospectively. By the nature of these *post hoc* defined analyses, these analyses could only be performed on an exploratory level without adjustments for multiplicity and are thereby from a formal perspective not suitable to establish statistical significance. The estimated effect size with the random effects meta-analysis was modest and while the nominal 95%-CIs excluded '0', the nominal p-values – while low - did not even fully achieve the equivalent evidence level that is usually required in a standard (two-pivotal trial) case. Against this background, also considering unavoidable uncertainties inherent in post-hoc analyses, and considering the proximity of the CI-boundaries to 'no effect', while the results could stem form a very small treatment effect, but this is not established based on usual standards.

Furthermore, the emphasis is put on the meta-analysis results pooling the two dose arms, which was not described in the analysis pans of the individual studies, and anyway the results for the pooled doses is not considered relevant for the assessment of the individual doses. The argumentation that the two doses offer similar benefit is also in contradiction with the posology as presented in the SmPC, where both doses are proposed separately. These aspects hamper the interpretation of the results.

The clinical relevance of the results is also questioned. The overall effect size (27 and 28 minutes less OFF time for 20 mg/day and 40 mg/day respectively) is considered meagre and not near the conventional minimally MCID of 1 hour. Also, the lower limit of the 95% CI is considered close to the null (7 and 9 minutes), well below the conventional 1 hour and also below the 20-30 minutes that the applicant concluded as MCID based on expert opinion and patient surveys results that cannot readily be extrapolated to the populations in the pivotal studies. Of note, the upper limit of the 95% CI (45 and 48 minutes) is not above the conventional MCID of 1 hour either. An aspect that was highlighted during the SAG-N where the experts agreed by consensus that the overall effect size was not considered clinically relevant.

As per the increase in ON time without troublesome dyskinesia, only two trials showed nominally significant results. However, these results are considered descriptive as statistically significance cannot be claimed due to lack of control for multiplicity. Additionally, in order to conclude on treatment benefit, significant changes in the OFF-time variable have to be coupled with significant improvements in ON time without troublesome dyskinesia. For both 20mg and 40mg doses the results are inconclusive for benefit in the 2 part responder analysis for the meta-analysis for the pivotal studies using random effects.

There was substantial heterogeneity across studies in the responder analyses as submitted during assessment. Notably, when viewing the responder analysis in Study 014 separately, a study conducted in subjects "optimally treated", the results showed that the percentage of responders was higher in placebo (39%) than in the two istradefylline arms (20 mg: 35%, 40 mg: 34%). This is a cause of concern as it suggests that in an optimally treated population there is no net benefit of adding istradefylline, but rather possibly a worse overall control of changes in OFF time coupled with good ON, as noted by the experts in the SAG-N.

As per Studies 007 and 014 enrolling European populations, the arguments and post-hoc analysis provided by the applicant as part of the re-examination are not convincing. Additionally, the *post-hoc* analysis cannot be accepted as pivotal evidence after having inconsistent or negative results in the primary and secondary analyses. Therefore, the fact that Studies 007 and 014 (that enrolled the European population) both clearly failed remains an unresolved issue.

It remains unclear whether there is any impact of race/ethnicity on efficacy of istradefylline treatment. However, it is noted that more favourable results were seen in both Japanese studies, and this again raises the concern on external validity and generalisability to the European population. During the SAG-N, experts agreed that the lack of positive results in studies including EU population was a concern.

There has also been no clear rationale for the dosing strategy of both 20mg and 40mg with no clear instruction for prescribers as to when to use a particular dose. In the 4 trials that studied both the 20 mg/ day and the 40 mg/ day dose, discordant results were observed. A trend towards a greater effect in reduction of OFF time and increase in ON time without troublesome dyskinesia for 40 mg vs 20 mg/ day was observed in study 018 (that was a negative study, showing no difference vs placebo) and in Study 0608 (Japanese study). Conversely, no difference between 20 and 40 mg/ day dose was observed in reduction of OFF time and increase in ON time without troublesome dyskinesia, in the other Japanese study (0609) and in Study 014.

Furthermore, the pooled results for 8 RCTs did not show relevant differences between the 20 and the 40 mg dose in total OFF (-0.38hr vs -0.45hr) nor in ON with non-troublesome dyskinesia (0.25hr for both doses). This further increases the overall uncertainty with regard to the observed efficacy and lack of efficacy in the different pivotal studies.

6.4. Unfavourable effects

Pool 1 includes 8 DB, randomised, placebo-controlled fixed-dose studies of 12- or 16-weeks duration (n=2073; 20 or 40 mg/day). Pool 2 includes 5 open label, single arm, long-term extension studies of istradefylline in subjects with idiopathic PD and motor response complications while taking levodopa therapy (n=2132 subjects).

TEAEs were reported more frequently in patients receiving istradefylline treatment. In Pool 1, 72.4% of subjects in the total istradefylline group experienced at least 1 TEAE compared to 65.4% of subjects in the placebo group. In Pool 2 89.7% of subjects reported any TEAEs.

Dyskinesia was the most commonly reported TEAE. In Pool 1 the incidence of dyskinesia was greater in the total istradefylline group (17.8%) compared with the placebo group (9.6%), with similar incidences of dyskinesia in the istradefylline 20 mg/day (16.1%) and istradefylline 40 mg/day (17.7%) groups. In addition, dyskinesia was the most frequently reported severe TEAE and the most frequently reported TEAE leading to discontinuation although the number of these patients was small. For other TEAEs reported with the higher frequency in the istradefylline group the difference as compared to placebo groups was smaller (1-2%).

The number of patients who experienced serious TEAEs in Pool 1 was small however, the percentage of such patients was slightly higher in the istradefylline group (4.2%) as compared to the placebo group (3.1%). More serious TEAEs were reported in patients receiving 40 mg dose as compared to those treated with 20 mg. The most frequently reported serious TEAEs were falls and pneumonia and for those there was no significant differences between istradefylline and placebo. Cardiac failure congestive and delirium were reported in 3 patients treated with istradefylline versus 0 cases in the placebo group.

Nine subjects died in Pool 1 and thirty-two subjects died in Pool 2 however, the provided data do not indicate that there is a higher risk of death of patients receiving treatment with istradefylline.

In Pool 1 TEAEs within the SOC Psychiatric disorders were reported more frequently in the istradefylline group (16.2%) as compared to those receiving placebo (11.4%). The highest frequency was seen in the highest dose group (24.5% in patients receiving 60 mg dose). TEAEs Anxiety had a slightly higher frequency in Pool 1 and in Pool 3.

Also serious TEAEs within the SOC Psychiatric disorders were reported with the higher frequency in the istradefylline group (10 cases in total) as compared to 1 case in the placebo group. The following serious TEAEs were reported: confusion sate, delirium, psychotic disorders, depression, disorientation, hallucination, persecutory delusion, psychiatric symptoms, suicidal attempt. It is noted that patients with psychotic illness or depression were excluded from all studies. Psychotic disorders including mania,

agitation, delirium, and abnormal behaviour, with some of these that considered serious, were reported only in patients treated with the proposed doses of istradefylline versus no case in the placebo group. The AE hallucinations was proposed by the applicant to be listed as ADR.

The slightly higher frequency of the upper respiratory tract infections observed in Pool 1 is apparently mainly driven by upper respiratory tract inflammation term. Respiratory TEAEs were the most frequently reported also in the long-term open-label Pool 2. In combination with the biological plausibility related to the istradefylline mechanism of action, the applicant has added upper respiratory tract inflammation as an ADRs.

The higher frequency of decreased appetite was observed in patients receiving treatment with istradefylline and that decrease in body weight was observed in the majority of patients enrolled to long-term studies.

Although the differences in incidence between istradefylline and placebo were small, all cases of impulse control disorder (ICD)(n=13) occurred in istradefylline-treated subjects. No serious ICD was reported, and discontinuation for ICD was observed only in one patient. Overall, 11 out of 13 subjects who experienced ICDs were on concomitant treatment with DAs. In the open-label long-term Pool 2, the majority of subjects with ICD were on concomitant DA treatment and resolved while istradefylline was continued.

Although sleep disorders are common in patients with PD, insomnia and abnormal dreams were reported more frequently in the istradefylline group compared with placebo. Insomnia and Vivid dreams are now included in the ADR Table of section 4.8 with the frequency "uncommon" based on the Pool 1 data.

Blood glucose increase was among the AEs that occurred in Pool 1 with a higher frequency compared with placebo. Therefore, blood glucose increase was requested to be considered as an ADR (similar as in the FDA label).

In non-clinical embryo-foetal development studies in rat fetotoxicity was evident at the high dose, with exposure margins likely to be < 3-fold. In rabbit, istradefylline was shown to be teratogenic with small exposure margins.

As of 31 May 2019, the cumulative post-marketing exposure estimate for istradefylline in Japan was approximately 63,500 patients. The most frequently reported AEs in the post-marketing setting, dyskinesia, hallucinations, constipation, dizziness, nausea and vomiting are recognized as adverse reactions with istradefylline based on clinical trial data. The most commonly reported SAEs were pneumonia and pneumonia aspiration. During routine signal detection activities, a safety signal of rash with istradefylline was detected from post-marketing data in Japan.

6.5. Uncertainties and limitations about unfavourable effects

There are concerns that the higher proposed dose of istradefylline (40 mg/day) could be slightly less safe as compared to the lower dose especially in relation to the development of serious and severe TEAEs after a long-term treatment with istradefylline.

There are still some uncertainties in relation to the development of foci of vascular mineralization which was seen in preclinical studies. The applicant claims that the available safety data provide reassurance that this concern is not relevant to humans. This is not fully agreed. The applicant agreed to add brain vascular mineralisation to the RMP as an important potential risk subsequently re-named as 'Movement, neurological or psychiatric disorders due to brain vascular mineralisation'. It was considered however that this important potential risk could be further re-named as follows: "Neurological (mainly movement) disorders or psychiatric disorders which the applicant agreed on.

A further discussion was required in relation to the potential risk of cardiac disorders taking into consideration the mechanism of action of istradefylline, which through its antagonistic action on the A_{2A} receptors could mediate coronary artery vasoconstriction, increase heart rate and blood pressure. Cases of cardiac SAEs were presented by the applicant which were considered as possibly related to istradefylline. The applicant was requested to commit monitoring of any signal on cardiac events suggestive of potential causal relationship with istradefylline treatment in the post-marketing setting.

The safety data on istradefylline for patients over 85 years of age are very limited and, hence, definitive conclusions on safety in this subpopulation cannot be made.

6.6. Effects Table

Table 145: Effects Table for istradefylline 20mg and 40mg as adjunctive treatment to levodopa

Effect	Short Description	Unit	Treatment		Control	Uncertainties / Strength of evidence	References
Favourable Effects							
Positive Studies			**Placebo**	**istradefylline 20 mg**	**istradefylline 40 mg**		
Study US-005 Reduction in OFF Time	Primary endpoint % change from baseline to endpoint in awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	N=66 -3.71%	N/A	N=130 -10.49% -6.78% -11.63, -1.92 0.007	Same effect size not replicated consistently in other studies Confidence intervals are wide Conducted only in North America, >15 years ago	
Study US-013 Reduction in OFF Time	Primary endpoint % change from baseline to endpoint in awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	N=115 -4.92%	N=116 -9.49% -4.57% -8.55, -0.59 0.025	N/A	Conducted only in North America, >15 years ago Confidence intervals are wide	

Effect	Short Description	Unit	Treatment		Control	Uncertainties / Strength of evidence	References
Study 6002-0608	Primary endpoint Change from baseline to endpoint in HOURS awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	N=119 -0.66h	N=119 -1.31h -0.65h -1.23, -0.07 0.028	N=125 -1.58h -0.92h -1.49, -0.35 0.002	Conducted in Japan, greater proportion of female participants Same effect size not replicated consistently in other studies	
Study 6002-009	Primary endpoint Change from baseline to endpoint in HOURS awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	N=126 -0.23h	N=123 -0.99h -0.76h -1.30, -0.22 0.006	N=124 -0.96h -0.74h -1.27, -0.20 0.008	Conducted in Japan, greater proportion of female participants Same effect size not replicated consistently in other studies No difference between the 20mg and 40mg doses	
Negative Studies (included considering there were pivotal studies)			**Placebo**	**istradefylline 20mg**	**istradefylline 40mg /(60mg)***		
Study US-006 Reduction in OFF Time	<u>Primary endpoint</u> % change from baseline to endpoint in awake time spent in OFF state	LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	N=77 -4.07% ANOVA	N=163 -7.72% -3.65% -7.83, 0.53 0.088	(60 mg) N=155 -7.84% -3.77% -8.01, 0.47 0.082	Prespecified analysis was ANOVA, however also performed ANCOVA which showed nominal statistical significance No difference between 20mg and 60mg doses Conducted only in North America, >15 years ago	

Effect	Short Description	Unit	Treatment		Control	Uncertainties / Strength of evidence	References
Study US-018	Primary endpoint % change from baseline to endpoint in awake time spent in OFF state	 LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	N=154 -7.59%	(20 mg) N=149 -6.11% 1.50% -2.05,5.05 0.408	(40 mg) N=152 -9.08% -0.66% -4.21, 2.88 0.714	Statistical significance not reached. Istradefylline 10mg dose also included. Istradefylline 10mg and 20mg were both inferior to placebo	
Study EU-007	Primary endpoint % change from baseline to endpoint in awake time spent in OFF state	 LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	Placebo N=152 -4.53%	**Entacapone 200mg** N-=153 -5.14% -3.29% -6.77, 0.19 0.064	Istra 40mg N=159 -7.82% -0.61% -4.05, 2.83 0.729	Did not meet statistical significance for Istra 40mg in European population and numerically Entacapone active comparator performed better, also when comparing CI.	
Study US-014	Primary endpoint Change from baseline to endpoint in HOURS awake time spent in OFF state MMRM	 LSM change from baseline to endpoint LSM difference from placebo in change from baseline to endpoint 95% CI p-value	Placebo N=204 -0.88 h	Istra 20mg N=202 -1.20 h -0.32 h -0.76, 0.12 0.156	Istra 40mg N=207 -1.15 h -0.27 h -0.70, 0.17 0.234	Failed to show a benefit in a more 'severe' population required to have dyskinesias and be on >/=400mg levodopa/day No difference between 20mg and 40mg doses	

Effect	Short Description	Unit	Treatment		Control	Uncertainties / Strength of evidence	References
Pooled Analysis of 8 RCTs Pool E1	Total HOURS OFF	LSM change from baseline to endpoint	Placebo	Istra 20mg	Istra 40mg	Effect size not compelling and clinical relevance is questioned for OFF time and ON time without troublesome dyskinesias No evident difference in treatment effect between 20mg and 40mg istradefylline doses	Table 2.7.3-17 SCE
		LSM difference from placebo in change from baseline to endpoint 95% CI		-0.38 -0.61, -0.15	-0.45 -0.68, -0.22		
	Total HOURS ON without troublesome dyskinesia	p-value		0.40	0.33		
		LSM difference placebo in change from baseline to endpoint 95% CI p-value		0.15, 0.66	0.08, 0.59		
Unfavourable Effects							
	Incidence of Dyskinesia %	%	17.2		9.2		
	Incidence of Nausea	%	5.9		3.7		
	Incidence of Dizziness	%	4.4		3.4		
	Incidence of Constipation	%	4.1		2.5		

6.7. Benefit-risk assessment and discussion

6.7.1. Importance of favourable and unfavourable effects

The main problem of this application is the borderline and inconsistent effect that istradefylline has on PD patients in the different trials submitted despite they have quite similar design. This raises doubts on the reliability, relevance and representativeness of the outcome of positive trials. The results are particularly poor for the European population which was studied in the more recent trials of the clinical development. Such studies would better reflect the current therapeutic approach to PD. Considering the overall weak studies results, a clinical effect of istradefylline cannot be concluded.

The original application contains eight pivotal clinical studies, with four positive studies and four negative studies (thereof 3 out of 6 for both doses). The weak estimates and inconsistency in effect conclusions between the studies does not allow to conclude on a positive benefit/risk.

The only two clinical studies that succeeded in showing a concurrent improvement with istradefylline treatment on both OFF and ON states, have the weakness that the selection of the ON endpoint was done *post hoc* or with no adjustment for multiplicity. Further limitations of the evidence supporting treatment benefit, come from the lack of confirmation of treatment efficacy in improving motor function,

(by UPDRS III score), and ameliorating clinical global status of patients (as per CGI scale) in the two positive trials in Caucasian patients.

It also needs to be borne in mind that the positive US studies were conducted over 15 years ago and given the availability of newer therapies, the study results may not be generalizable to a present day European population, especially considering the two studies which enrolled European patients were negative studies, 6002-EU-007 and 6002-014.

Given the inconsistency of the individual study results, the applicant undertook a pooled analysis of the 8 pivotal studies. The pooled analysis had not been predefined, but was rather performed as a rescue measure following the observation that the individual trials, which were planned to stand on their own, showed only borderline or no effect. Next to the lack of pre-definition of the meta-analysis as performed, there were other methodological constrains, e.g. there was notable heterogeneity between studies and the absence of statistical rigour in terms of that the nominal p-values, that did not achieve the equivalent to the evidence level that is applied in a (two-pivotal trial) standard case, also considering additional uncertainties inherent in the post-hoc analyses. Also the estimated effect size with the random effects meta-analysis was modest and the lower limits of these unadjusted CIs correspond to 9min resp. 7.2min only. Therefore, while the 95%-confidence interval excluded '0', given the proximity of the CI-boundaries to 'no effect', a clinically relevant treatment effect cannot be confirmed.

These methodological considerations preclude the use of the meta-analysis as pivotal evidence for the MAA. Even these issues related to the meta-analysis could be deemed acceptable, the poor clinical relevance of the results remains. The overall effect size (27 and 28 minutes less OFF time for 20 mg/day and 40 mg/day respectively) is considered meagre and not near the conventional MCID of 1 hour. In the SAG-N, experts agreed by consensus that the estimated effect size could not be condisered as clinically relevant. The cut-off of 1 hour was also mentioned as threshold for a MCID in the meeting and it was noted that not even the upper limit of the 95%CI was above 1 hour. Further, as per the increase in ON time without troublesome dyskinesia, only two trials showed nominally significant results, meaning that there is no unequivocal support from an important secondary endpoint.

In the responses to the grounds for refusal, the applicant has provided a number of analyses of the changes in OFF and ON time to support the beneficial effect of istradefylline in the European population. However, such benefit cannot be considered demonstrated. The analysis in maximally treated patients shows slight differences versus placebo, and the region analysis only increases the concerns regarding a potential effect of istradefylline in the European population as the effect in OFF and ON time seem to be essentially driven by the Japanese subgroup.

The applicant has tried to justify the effect size in OFF and ON time providing the results from some patient surveys conducted in some European countries. The applicant proposes from these that a decrease of 20-30 minutes in OFF time or similar increase in ON time without troublesome dyskinesia, is relevant by patients. However, such interviews are not free of bias considering how questions were formulated. Furthermore, the baseline time in OFF state is key to understand the relevance of the gained time. While patients in the surveys had a daily duration in OFF time less than 1 hour, the mean number of hours spent in OFF state in the trials ranged from 5.36 to 6.7 hours/day, making these results hardly generalizable. In any case, while patient perceptions may be useful as supportive information they cannot replace the need of robust data from clinical trials and cannot be seen in isolation from the inconsistency of and borderline studies results.

While it can be agreed that the safety profile could be considered manageable with the appropriate risk minimization measures, several adverse-drug reactions were identified for Nouryant. Additionally, cases of cardiac SAEs were presented by the applicant which were considered as possibly related to istradefylline and the applicant was requested to commit monitoring of any signal on cardiac events suggestive of potential causal relationship with istradefylline treatment in the post-marketing setting.

Finally, the uncertainties about the long-term safety profile in particular given the absence of evidence from the dose findings studies supporting the proposed dose of 40mg cannot be ignored.

6.7.2. Balance of benefits and risks

Replication of clinical results from trials with similar designs and including similar populations is an important notion since this provides confirmation of the true effect of a given drug. Likewise, the lack of consistency questions the reliability of the positive studies. In this particular case, despite four positive trials that met the primary efficacy endpoint, there are four formally negative trials where clinical benefit of istradefylline has not been shown. The degree of inconsistency despite similarly designed clinical studies raises concerns over the potential for this drug to provide benefit to patients.

Efficacy cannot be considered established based on the eight individual studies due to inconsistent results nor on the provided descriptive meta-analyses results, now provided as pivotal evidence. Even if the methodological constraints would be put aside, the results of the meta-analyses are not deemed of clinical relevance.

Importantly, half of the studies including those enrolling European patients and the most recent trial which enrolled a theoretically more severe PD population have not demonstrated any evidence of clinical benefit with addition of istradefylline to ongoing PD treatment. Analysis in maximally treated patients only shows slight differences versus placebo. Even for the studies in which some efficacy has been shown, both Japanese studies, the higher proportion of females makes generalisation to European population difficult, and the clinical relevance of any observed treatment effect size is questioned.

There has also been no clear rationale for the dosing strategy of both 20mg and 40mg. The studies did not demonstrate any additional benefit for the 40mg over the 20mg Istradefylline dose, and no dose dependency has been shown between the doses.

Finally, the surveys/interviews to patients and prescribers submitted by the applicant to support the effect size of the increase in OFF time can only be considered as complementary to data from clinical trials but they can never replace the need of robust and consistent positive results from adequately designed clinical trials.

So, while the safety profile is not negligible for the target population and would not be prohibitive for approval, the B/R is negative as efficacy remains not established.

6.7.3. Additional considerations on the benefit-risk balance

Not applicable

6.8. Conclusions

The overall B/R of Nouryant is negative.

7. Recommendations following re-examination

Based on the arguments of the applicant and all the supporting data on quality, safety and efficacy, the CHMP re-examined its initial opinion and in its final opinion concluded by consensus that the efficacy of Nouryant is not sufficiently demonstrated, and, therefore recommends the refusal of the granting of the marketing authorisation for the above mentioned medicinal product. The CHMP considers that:

The applicant submitted eight pivotal clinical studies in support of this application. While four of the eight pivotal trials submitted met their primary endpoint of a reduction in the % change from baseline to endpoint in time spent OFF (studies US-005, US-013) or change from baseline to endpoint in total hours spent OFF (Japanese studies 0608 and 009), the other trials did not.

Therefore, the efficacy of the product is not demonstrated considering the inconsistency of the results across the development program. More specifically:

- Study US-006 was a formally failed study as per the pre-specified primary ANOVA analysis model. The primary analysis model was changed from ANOVA to ANCOVA after finalising the SAP. Considering the predefined statistical analysis, this study is not considered to have demonstrated a statistically significant treatment effect.
- The inconsistency of study results including the different responses observed in the studies in different regions and the difference in response over time, are not resolved by post-hoc pooling the individual trials. In addition, the estimated modest effect size, and the unadjusted nominal confidence interval that is in close proximity to no effect are not considered sufficient to have demonstrated a treatment effect in a post-hoc pooled analysis. Moreover the observation of no clear pattern of a dose response with increasing doses of istradefylline, leads to considerable uncertainty on the effect of the treatment.
- The fact that Studies 007 and 014 (that enrolled the European population) both clearly failed remains an unresolved issue. In particular, as the most recent study, Study 6002-014 in 'maximally and optimally treated' patients did not show positive results – despite being the largest pivotal trial relative to size of each treatment group and despite being planned taking into account the results of previously conducted istradefylline's trials.

The CHMP is of the opinion that the efficacy of the above-mentioned medicinal product is not properly, or sufficiently demonstrated. Therefore, the CHMP has recommended the refusal of the granting of the marketing authorisation for Nouryant.

Furthermore, following review of the available data in the context of the applicant's claim of new active substance status, the CHMP position at the time of this report is reflected in section 2.9 (new active substance). However, in light of the negative recommendation, the CHMP is of the opinion that it is not appropriate to conclude on the new active substance status at this time.