SCIENTIFIC DISCUSSION

1. Introduction

AZILECT is indicated for the treatment of idiopathic Parkinson's disease (PD) as monotherapy (without levodopa) or as adjunct therapy (with levodopa) in patients with end of dose fluctuations. Rasagiline is administered orally, at a dose of 1 mg once daily with or without levodopa.

Parkinson's disease is a common neurodegenerative disorder typified by loss of dopaminergic neurones from the basal ganglia, and by a characteristic clinical syndrome with cardinal physical signs of resting tremor, bradikinesia and rigidity. The main treatment aims at alleviating symptoms through a balance of anti-cholinergic and dopaminergic drugs. Parkinson's disease (PD) treatment is complex due to the progressive nature of the disease, and the array of motor and non-motor features combined with early and late side effects associated with therapeutic interventions.

Rasagiline is a chemical inhibitor of the enzyme monoamine oxidase (MAO) type B which has a major role in the inactivation of biogenic and diet-derived amines in the central nervous system. MAO has two isozymes (types A and B) and type B is responsible for metabolising dopamine in the central nervous system; as dopamine deficiency is the main contributing factor to the clinical manifestations of Parkinson's disease, inhibition of MAO-B should tend to restore dopamine levels towards normal values and this improve the condition. Rasagiline was developed for the symptomatic treatment of Parkinson's disease both as monotherapy in early disease and as adjunct therapy to levodopa + aminoacids decarboxylase inhibitor (LD + ADI) in patients with motor fluctuations.

2. Quality

Introduction

Drug Substance

- Composition

AZILECT contains rasagiline mesylate as the active substance. It is presented as tablets each one containing an amount of rasagiline mesylate equivalent to 1 mg of rasagiline base

Other ingredients include mannitol, maize starch, pregelatinized maize starch, colloidal anhydrous silica, stearic acid and talc.

AZILECT is packaged in high density polyethylene (HDPE) containers or in Aluminium (Alu/Alu) blister packs.

- Active substance

The chemical name of rasagiline mesylate is N-propargyl-1(R)-aminoindan-mesylate. It is a white to off-white powder, freely soluble in water or ethanol and sparingly soluble in isopropanol. Rasagiline is a highly soluble substance, according to the Biopharmaceutics Classification System. Rasagiline mesylate is a chiral compound with one asymmetric carbon atom in the five-member ring with an absolute R configuration, which is produced as a single enantiomer. The proof of the chemical structure of rasagiline mesylate drug substance has been achieved by elemental analysis, UV, IR, ^{1}H- and ^{13}C-NMR spectroscopic methods and MS. All data were consistent with the proposed structure. There is no evidence of polymorphism and only one crystalline form of rasagiline mesylate exists.

The drug substance synthesis is carried out in 3 steps starting from the main starting material 1-aminoindan. The route of synthesis has been adequately described and is appropriately controlled with suitable in-process controls.

Validation results from three production-scale validation batches using the commercial route of synthesis demonstrated that the active substance is produced in a reproducible manner and that it meets consistently the proposed specifications.

The potential organic impurities of rasagiline mesylate drug substance originating from the route of synthesis as well as from the degradation of the active ingredient have been thoroughly discussed.

Only 5 of these have been detected in batches of the active substance and specifications have been set for them. No heavy metals are used throughout the synthesis of rasagiline mesylate and the content of residual organic solvents in rasagiline mesylate is in accordance with the ICH guideline Q3C requirements.

Active substance specification

The active substance specification includes tests for description, identification (IR, UV and HPLC), colour and clarity, melting point, specific optical rotation as indicative of the chirality of the material, loss on drying, sulphated ash, heavy metals, pH of an aqueous solution, assay, impurities and degradation products (HPLC, GC-MS), enantiomeric purity, organic residual solvents, microbial limit and particle size distribution.

The proposed specifications for the drug substance are based on the analysis of 15 batches of rasagiline mesylate. The analytical methods used in the routine controls have been suitably described and validated, while the limits for organic impurities have been qualified and accepted based on the levels observed in toxicological studies.

Batch analysis data from 15 batches have been presented. All batches comply with the active substance specification.

Stability

Stability studies were carried according to ICH requirements on 3 primary stability batches of rasagiline mesylate and 3 supporting production batches. Samples were stored at 25 °C/60%RH for up to 48 months and at 40 °C/75%RH for 6 months.

Stress testing was also carried out, which included photostability studies performed under ICH conditions and a series of forced degradation studies designed to determine the conditions under which degradation occurs and to assess the stability indicating ability of the analytical methods used for related substances.

The parameters evaluated during the stability studies were description, assay (HPLC, titration), impurities/degradation products determination and enantiomeric purity.

No significant changes have occurred in any test parameter under normal storage conditions. The results of all stability studies indicate that rasagiline mesylate is stable and that the container closure system that was used provides suitable protection for the drug substance under normal storage conditions.

- Other ingredients

All excipients used in the product are of non-animal origin and comply with their corresponding European Pharmacopoeia monographs.

The immediate packaging materials are commonly used for tablets and consist of either HDPE bottles with a polypropylene cap or (Alu/Alu) blisters.

Drug Product

Product development and finished product

The pharmaceutical development was aimed at obtaining an immediate-release tablet. The mesylate salt was selected as the drug substance for the commercial product for its stability. Preformulation studies with various excipients led to the selection of mannitol as the main filler pregelatinized maize starch and starch as binder and disintegrant, talc and stearic acid as lubricants and colloidal silicon dioxide as glidant for the final formulation.

The main objective of the manufacturing process development was to obtain a medicinal product that presents good uniformity of content, dissolution and other critical physical parameters. For this reason a standard granulation process was employed comprising of the following steps: mixing, granulating drying and milling. The resulting granules are compressed into tablets and then packaged. All critical process parameters have been identified and controlled by appropriate in process controls. The validation report from three production scale batches demonstrates that the process is reproducible and provides a drug product that complies with the in-process and finished product specifications.

Different product formulations have been used in the early stability studies. However bioequivalence between the clinical trial formulations and the one intended for marketing has been demonstrated and is further supported by the results of an in-vivo pharmacokinetics study.

Product specification

The specification for the finished product at release and shelf life includes tests for description, identification (TLC and HPLC), assay, uniformity of content, dissolution, weight, thickness, hardness, friability, disintegration, impurities and degradation products (HPLC and GC-MS), and microbial count. All tests included in the specification have been satisfactorily described and validated.

Batch analysis data from 7 clinical and 6 industrial batches have been presented. All batches met the test limits as defined in the release specification and test methodology valid at the time of batch release.

Stability of the product

Stability studies were carried out on 3 production scale batches of tablets according to the ICH requirements. Batches were packaged in Alu/Alu blisters and HDPE bottles capped with polypropylene caps. Samples were stored at 25°C/60 % RH for up to 24 months, 30°C/60 % RH for up to 12 months and 40°C/75 % RH for 6 months.

The parameters tested concerned physical and chemical testing of the same quantifiable parameters as for release i.e. assay/Identification (HPLC), identification of impurities, dissolution, hardness, microbial testing. The stability results presented were satisfactory and support the proposed shelf life for the commercially packaged product under the conditions specified in the SPC.

Additional photostability studies have been performed according to ICH guidelines in both packages and the results demonstrated that the product is also stable to light.

Discussion on chemical, pharmaceutical and biological aspects.

The quality of AZILECT is adequately established. In general, satisfactory chemical and pharmaceutical documentation has been submitted for marketing authorization. There are no major deviations from EU and ICH requirements.

The active substance is a highly soluble substance, according to the Biopharmaceutics Classification System. It is well characterised and documented. The excipients are commonly used in these types of formulations and comply with Eur. Phar. requirements. The packaging material is commonly used and well documented. The manufacturing process of the finished product is a standard granulation process that has been adequately described. Stability tests indicate that the product under ICH guidelines conditions is chemically stable for the proposed shelf life.

3. Non-Clinial aspects

Introduction

AZILECT is an oral tablet containing 1 mg of rasagiline expressed in terms of rasagiline base, which has a molecular weight of 171.23. The recommended posology is once daily oral administration of a 1.0 mg Tablet.

Two salts were used during the non-clinical programme. Initially, the hydrochloride salt was used, but this was changed to the mesylate salt to overcome stability issues. Bridging studies have been conducted, and bioequivalence has been demonstrated.

There was no bioconversion of rasagiline mesylate (R enantiomer) to its S enantiomer within the human body, as determined in plasma samples for healthy volunteers dosed with rasagiline.

Nonclinical studies with rasagiline were conducted in mice, rats, rabbits, dogs and marmosets. Sufficient exposure multiples were achieved *in vivo* to allow comparison with clinical data, and in the calculation of safety margins.

Pivotal toxicology studies were stated to be conducted to GLP standards.

Pharmacology

- Primary pharmacodynamics (in vitro/in vivo)

Monoamine oxidase (MAO) plays a major role in the in vivo inactivation of biogenic and diet-derived amines in both the central nervous system (CNS) and in peripheral neurons and tissues. Two MAO isozymes are distinguished:

- MAO type A (MAO-A), which is mainly responsible for the deamination of serotonin (5-hydroxytryptamine; 5-HT) and noradrenaline (NA). Dopamine (DA) is a substrate for both MAO-A and MAO-B in the brain. In the intestine, MAO-A metabolizes the oxidation of tyramine.
- MAO-B, which is inhibited by selegiline. MAO-B inhibition in the human brain principally reduces the catabolism of DA and β-phenylethylamine (PE).

Both MAO-A and MAO-B are found in similar amounts in rodent brains, whereas in man, MAO-B predominates (80% of total MAO) in the brain and MAO-A is found in greater abundance in the gastrointestinal tract.

Inhibition of MAO activity by rasagiline [N-propargyl-1(R)-aminoindan] mesylate, rasagiline hydrochloride, their (S)-isomers and by the metabolite 1-(R)-aminoindan hydrochloride was studied *in vitro* and *ex vivo*. The MAO inhibitory properties of rasagiline were compared to those of the selective irreversible MAO-B inhibitor selegiline.

Both rasagiline mesylate and rasagiline hydrochloride exhibit a similar, highly potent and MAO-B - selective inhibition activity *in vitro*. The derived IC50 values for human and rat brain MAO-B inhibition are 20-90 fold lower compared to the respective values for MAO-A inhibition. The metabolite of rasagiline, 1-R aminoindan, did not inhibit brain MAO-B *in vitro*.

The high potency and relative selectivity of rasagiline seen in brain and peripheral tissues *in vitro* was preserved *in vivo* following oral acute administration in rodents. Rasagiline was more potent than selegiline. The ratio of their respective ED50 values for inhibiting rat brain or liver MAO-B are 1/3 or 1/13, respectively.

In studies in rats for 21 days (po), repeated treatment with rasagiline resulted in increased MAO (A and B) inhibition compared to acute treatment. Rasagiline selectivity for MAO-B inhibition was maintained upon repeated treatment. The superior MAO inhibitory potency of rasagiline over selegiline was maintained also in repeated treatment. Rasagiline exhibited selectivity for brain and peripheral tissues MAO-B inhibition also in dogs. Rasagiline was also shown to be a potent inhibitor of the marmoset brain MAO-B.

To demonstrate the irreversible nature of MAO inhibition by rasagiline, recovery of MAO activity following chronic drug administration was monitored. Seven and 13 days after cessation of treatment brain, MAO-B and –A are still inhibited by 55-40% and 45-41%, respectively. The irreversibility of brain MAO inhibition was also established in marmosets.

In studies in mice, both rasagiline and selegiline prevented the 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP)-induced decline in DA level in a dose-dependent manner. However, the minimal rasagiline dose needed to block 100% of the MPTP-induced damage was 0.5 mg/kg compared to a selegiline dose of 2.5 mg/kg. In marmoset, treatment with either rasagiline or selegiline (10 mg/kg, s.c.) also markedly attenuated the neurotoxic effects of MPTP.

Increases in DA and 5-HT level occurred at rasagiline doses of 2 and 5 mg/kg/day, at which MAO-A was inhibited to a large extent. These increases in monoamine levels were associated with decreases in the levels of the respective metabolites, indicating that the changes in monoamine levels were the direct consequence of MAO-A and MAO-B inhibition. Studies which used the microdialysis method to determine monoamine levels showed that both rasagiline and selegiline caused an increased extracellular DA level following chronic treatment in the rat, at a dose, which caused selective MAO-B inhibition.

Rasagiline restored normal motor activity in animal models of haloperidol or α-MpT -induced dopaminergic dysfunction. Acute pre-treatment with rasagiline (0.5-6 mg/kg, i.p.) prevented haloperidol-induced catalepsy and α-MpT-induced hypokinesia in adult rats.

The major rasagiline metabolite, 1(R)-aminoindan, exhibited ameliorating activity on α-MpT-induced hypokinesia, potentiated amphetamine-induced stereotypic behavior and improved performance in the passive avoidance test.

- Secondary pharmacodynamics

Rasagiline neuroprotective properties were studied *in vitro* in dopaminergic-mesencephalic, cortical and hippocampal primary neuronal cultures and in the PC12 and SH-SY5Y cell lines. The neuroprotective effects of rasagiline *in vitro* cannot be attributed in the main to MAO inhibition by rasagiline. This is because its (S)-enantiomer, which lacks MAO inhibitory activity, exhibited an almost equivalent protective activity in these cell culture systems. These pharmacological actions have been demonstrated to be associated with the propargylamine moiety, since propargylamine itself has similar action.

Rasagiline neuroprotective potential was confirmed in several *in vivo* models of neuronal insult: hypobaric hypoxia, closed head injury in mice, permanent focal ischemia in rats, stroke susceptibility in stroke-prone hypertensive rats, ALS-like syndrome in transgenic Cu/Zn SOD mice, apoptotic death of immature rat motor neurons following axotomy and 6-hydroxydopamine (6-OHDA) induced degeneration of substantia nigra dopaminergic neurons.

The rasagiline metabolite, aminoindan, given to animals, has exhibited ameliorating activity in several models of dopaminergic motor dysfunction and hypoxia-induced cognitive deficits.

- Safety pharmacology

The effect of rasagiline on the cardiovascular system was assessed in dogs, rats and cats. In the dog study, conscious animals acutely treated with rasagiline (3 mg/kg, oral) were monitored by telemetry and blood pressure, heart rate as well as ECG parameters were measured. Rasagiline produced no overt treatment related changes in cardiovascular parameters in dogs. Similarly, rasagiline (up to 1 mg/kg, i.v.) caused no effects on the cardiovascular system in cats.

Studies in rats indicated that rasagiline administered repeatedly at dose levels (2 mg/kg, oral) which exceeded those required for selective MAO-B inhibition showed no changes in cardiovascular parameters. In a comparative study that assessed the effects of rasagiline (acute 5 and 10 mg/kg, i.v.) versus selegiline (acute 5 and 10 mg/kg, i.v.) on blood pressure of pithed rats, only selegiline caused significant increases in blood pressure.

The tested doses of rasagiline in the rat and dog were at least 10 times higher compared to the clinical dose of 1 mg/day in patients. Therefore a satisfactory margin of safety was provided.

The effect of orally administered rasagiline mesylate on central nervous system functions governing motor activity, behavior and response to stimuli in the rat were monitored using the Irwin test. Rasagiline evoked no unusual effect in this test.

Respiratory adverse events were absent in a dog telemetry study and in repeated dose toxicology studies.

- Pharmacodynamic drug interactions

A potential pharmacodynamic interaction of rasagiline with Levodopa was considered. The effects of rasagiline versus selegiline on the Levodopa pressor response were compared in a series of studies. When rasagiline and selegiline had been given acutely (i.v.) to pithed rats at a dose of 5 mg/kg, acutely administered levodopa evoked a pressor response in the case of selegiline but not rasagiline. Levodopa co administered with carbidopa failed to evoke a pressor response after pre-treatment with rasagiline for 15 days (1 mg/kg/day, p.o.). Mean blood pressure decreased following repeated levodopa/carbidopa administration to rats. Mean BP in rasagiline-treated (1 mg/kg) rats given levodopa/carbidopa did not change significantly from those given saline and levodopa/carbidopa.

The effect of rasagiline on tyramine pressor response was studied in rats. Tyramine failed to evoke a pressor response after pre-treatment with rasagiline when given at relevant pharmacological doses (up to 0.5 mg/kg/day, p.o. for 21 days), at which MAO-A was only partially inhibited. (See Clinical section)

Pharmacokinetics

Toxicokinetic monitoring of systemic (plasma) exposure to unchanged product and the metabolite 1-aminoindan was incorporated into the design of the repeat-dose toxicity studies, carcinogenicity studies and reproduction toxicity studies. Studies were also undertaken in rats and dogs to investigate the potential for pharmacokinetic drug-interactions when rasagiline is co-administered with levodopa and the peripheral decarboxylase inhibitor carbidopa.

Non-clinical studies were conducted with the both the hydrochloride and mesylate salts of rasagiline. However, bioequivalence has been demonstrated, and the two salt forms can be considered as essentially the same.

^{14}C-labelled and unlabelled rasagiline and its metabolite aminoindan have been used in the quantification of PK parameters.

- Absorption- Bioavailability

Oral studies with ^{14}C-rasagiline show that absorption is rapid in all species, with Cmax attained in less than 2 hours. Absolute bioavailability has been estimated as 53-69% in rats, 13-22% in dogs, and 36% in humans.

Toxicokinetic analyses during the toxicology studies showed that exposure was linear at doses higher than the pharmacological selectivity for inhibition of MOA-B and was maintained up to about 5 mg/kg/day. However, kinetics became non-linear at higher doses, possibly indicating saturation of the elimination processes for both rasagiline and its metabolite aminoindan.

Accumulation was seen only at the highest doses in the mouse and dog studies (60 and 21mg/kg/day respectively).

- Distribution

IV studies in rats and dogs show that the volume of distribution (V_d) of rasagiline is several times that of total body water, indicating extensive tissue distribution.

Tissue distribution of ^{14}C-rasagiline was studied in albino and pigmented rats, revealing peaks of tissue radioactivity between 0.25 and 0.5 hours. Distribution to large intestine, urinary bladder and lacrimal glands takes longer, whilst persistence (up to 24 hrs) was seen in eyes, skin and arterial walls of pigmented animals.

In-vitro protein binding in plasma of animals is in the range of 70 to 90% and in human plasma in the range of 88 to 94%.

- Metabolism (*in vitro/in vivo*)

An extensive first pass metabolism effect is evident, likely due to rasagiline binding to MAO sites in the intestine prior to passing the liver.

Metabolism is rapid and extensive, with a similar profile in all tested species. The primary route of biotransformation is via N-dealkylation to form aminoindan and by hydroxylation to form 3-hydroxy-N-propargyl-1-aminoindan. Conjugation by sulphide or glucuronic acid occurs.

Microsomal studies indicate CYP1A2 as the primary metabolising isotype, but rasagiline is neither an inducer nor inhibitor of cytochrome p450. The metabolism of rasagiline under inhibition, induction of CYP1A2 or in presence of concomitant substrate to the enzyme has been addressed clinically.

- Excretion

The clearance of rasagiline is greater than the combined predicted renal and hepatic blood flow, suggesting extra hepatic, extra renal modes of elimination, possibly through non-reversible binding to target sites in the tissues. Mass balance studies conducted following oral administration in mice, rats and dogs showed a large proportion (>90%) of the administered radioactivity in the excreta, particularly urine (70-85%).

Despite the rapid elimination half-life for rasagiline, repeat doses were seen to give greater exposure than seen after single doses. The company have reasonably argued that this observation is compatible

with the mechanism of irreversible binding of rasagiline to MAO-B, leading to saturation of binding site. Excess unbound product appears in the plasma after the saturation threshold has been reached.

Toxicology

- Single dose toxicity

Single dose toxicity studies were conducted by the intravenous route in rats and by the oral route in mice, rats and dogs. Studies were initially conducted with the hydrochloride salt and subsequently with the more stable mesylate salt. Single dose bridging studies were undertaken which established the essentially similar toxicity and kinetics profiles of the hydrochloride and mesylate salts in rats and dogs. Mortalities were induced in rats by intravenous doses ≥69 mg/kg/day. Mortalities were induced in mice at oral doses of ≥206 mg/kg, in rats at oral doses ≥155 mg/kg and in dogs at oral doses of ≥84 mg/kg. Death was a result of the functional neuropharmacological changes that can be anticipated when excessive doses of a molecule capable of inhibiting the oxidation of biogenic amines are administered. The maximal non-lethal oral dose for rats and mice was about 100 mg/kg/day and the maximum tolerable dose (MTD) in dogs was 42 mg/kg. These doses represent considerable multiples of the recommended clinically relevant maximum dose of 1 mg/patient/day.

- Repeat dose toxicity (with toxicokinetics)

The rat and dog were selected for the conduct of repeat dose toxicity studies, both species having been shown to be pharmacologically responsive to rasagiline-induced inhibition of MAO-B. Repeat dose intravenous toxicity studies of 4-week duration were conducted at maximum doses of 3 mg/kg/day in rats and 5 mg/kg/day in dogs. Repeat dose oral toxicity studies of up to 26 week duration were conducted in rats employing doses spanning the range 0.14 to 17 mg/kg/day and of up to 52 week duration in dogs employing doses spanning 0.28 to 21.0 mg/kg/day.

When the multiples of the systemic exposures to rasagiline and aminoindan across the range of doses used for the repeat dose toxicity studies in rats and dogs are compared with the clinically relevant human exposure at the maximum recommended daily dose of 1 mg/day/patient, it is observed that the lowest doses used for the repeat dose toxicity studies afforded at least 7-fold the mean C_{max} and AUC last values for PAI (rasagiline) in humans receiving 1 mg/day. The highest doses used for the repeat dose toxicology studies afforded at least 250-fold C_{max} and AUC last values for PAI in humans receiving 1 mg/day.

After intravenous and oral dosing the principal manifestations of toxicity were related to the loss of selectivity for MAO-B (i.e. reduced food intake and weight gain and hyperactivity and/or aggression in rats). At the higher oral doses these findings were sometimes accompanied by increases in liver weight and changes in hepatocyte morphology in rats. The liver changes were consistent with changes observed in rats treated with hepatic microsomal enzyme inducers, there was however no evidence from studies that measured hepatic microsomal proteins to support this hypothesis. Suspected changes in thyroid and bladder morphology identified in the rat 13-week oral study were not corroborated by findings in either the 4-week or 26-week rat oral studies. The no adverse effect level (NOAEL) defined after 26 weeks treatment of rats and 52 weeks treatment of dogs was 5.1 mg/kg/day (for both rats and dogs). In terms of AUC last values, the animal NOAELs afford multiples of at least 15-fold with respect to exposure to PAI at the clinical dose of 1 mg/patient/day.

Studies were conducted to examine whether co-administration of rasagiline with levodopa and a peripheral decarboxylase inhibitor (carbidopa) can produce effects other than the expected dopaminergic actions. There were no effects in rats or dogs given rasagiline/carbidopa/levopda that could not be attributed to amplification of the effects of levodopa/carbidopa.

- Genotoxicity *in vitro* and *in vivo*

Rasagiline mesylate was not mutagenic in the bacteria reverse mutation (Ames) assay, with or without metabolic activation nor clastogenic when tested in the absence of metabolic activation. In the presence of metabolic activation, equivocal results were obtained at the highest tested doses.

According to the applicant, based on published literature, gentamycin enhances liver mediated cytotoxicity, by interacting with molecules such as pargyline (which is of the same class as rasagiline) at the ribosomal level (Mayne & Evans, 1988). When pargyline was tested in the chromosome aberration assay a similar effect of gentamycin was observed. Addition of glutathione reduced the

incidence of aberrations to control levels. Rasagiline seemed therefore to display slight genotoxicity in the system in presence of metabolic activation, which would be enhanced by the presence of gentamycin.

In the TK mouse lymphoma assay, rasagiline showed excessive cytotoxicity and evidence of clastogenicity in the presence of metabolic activation. The effect was greatly reduced by the addition of glutathione.

In view of the equivocal in vitro results, two types of in vivo genotoxicity studies were performed rather than one. One study tested micronuclei formation in polychromatic erythrocytes in the bone marrow of mice, and one study tested unscheduled DNA synthesis in liver cells of treated rats. Both studies demonstrated no mutagenic potential for rasagiline mesylate.

The combination of rasagiline with levodopa/carbidopa did not show genotoxic potential in vivo.

- Carcinogenicity (with toxicokinetics)

The carcinogenic potential of rasagiline was studied in two life-span studies in mice and rats, using doses chosen on the basis of pharmacodynamic and toxicological criteria rather than on the basis of systemic exposure ratios in animals and man, due to the equivocal genotoxicity identified in vitro.

Rasagiline was not carcinogenic in rats at exposure up to 84 times the human exposure at the recommended clinical dose, for male rats and up to 399 times in female rats. Microscopic examination identified foot lesions, characterized by chronic-active inflammation and ulceration of the epidermis. Urogenital tract inflammation characterized by acute inflammation and haemorrhage of the urinary bladder and prostate gland with dilatation of the kidney pelvis and tubules and tubular degeneration were found at significantly increased incidence in highest dosage male rats group (3.0 mg/kg/day). These pathological findings seen in rats treated chronically at high doses were explained by exaggerated sympathetic activities that relate to the primary action of the product.

In the mice study, the doses administered were 1, 15 and 45 mg/kg/day. Increased incidences of combined lung adenomas/adenocarcinomas were observed in both genders, with a lower safety margin (based on human and animal exposure) in males (rasagiline exposure ratios of mice, to humans treated with 1 mg/day (AUC mice/AUC humans) were 6, 213 and 1418 for males and 4, 144 and 419 for females dosed with 1, 15 and 45 mg/kg/day, respectively.). Historical control data showed a wide range of incidences of lung tumors (combined adenoma/carcinoma in males up to 46% and in females up to 28.6%). Although it is recognized that even at 45 mg/kg/day the observed incidence of combined bronchiolar/alveolar adenoma and/or carcinoma was only slightly above the testing laboratory's historical control range for females and was within the historical control range for males CD-1 mice, there was a clear dose-related increase (13/13; 11/12; 23/19; 24/20 respectively in male/female controls, 1, 15 and 45 mg/kg/day dose groups). Furthermore, safety margin at NOEL for males was only 6 (even if it should be noted that there is a 15 fold increase in between the low and intermediate doses). Considering the equivocal genotoxic potential and the lack of plausible explanation for lung tumor development in mice, the company was asked to perform further test addressing the involvement of a genotoxic mechanism in the lung tumorigenesis observed in mice. Results will be provided post-marketing.

- Reproductive and developmental studies

Studies were undertaken to assess the potential for rasagiline to adversely affect fertility and reproductive performance and peri- and postnatal- development in rats and embryo-foetal development in rats and rabbits. Studies of the potential to adversely affect embryo-foetal development by co-administration of rasagiline/levodopa/carbidopa were also undertaken in response to advice and comments received from US FDA reviewers.

Despite an evident dose-dependent lowering of serum prolactin, administration of rasagiline to male rats at doses of 0.5, 2.0 and 5.0 mg/kg/day from 28 days prior to cohabitation with untreated female rats did not affect the fertility or reproductive performance of male rats. Similarly there were no adverse effects on fertility and reproductive performance or embryo-foetal development in a study in which females were treated, from 14 days prior to cohabitation with untreated males until day 17 post coitum, at doses of 0.3, 1.0 and 3.0 mg/kg/day.

When oral doses of 1, 7, and 45 mg/kg/day were administered to female rabbits on days 6 to 20 post coitum, mean post-implantation losses were increased and mean foetal weight was reduced at the maternally toxic dose of 45 mg/kg/day. There were no other adverse effects on embryo foetal development.

There was an increased incidence of wavy ribs in rat foetuses obtained from dams that were treated with rasagiline/levodopa/carbidopa at ≥0.3/80/20 mg/kg/day on days 8 to 17 post coitum. No toxicological importance was attached to this finding since it is documented as a transient and reversible phenomenon which most probably results from disturbances of the materno-placental circulation.

Co-administration of rasagiline/levodopa/carbidopa at doses of 0.1/80/20, 0.6/80/20 or 1.2/80/20 on days 6 to 18 post coitum elicited maternal toxicity in rabbits with increased mean implantation losses at 0.6/80/20 and 1.2/80/20 mg/kg/day.

A pilot investigation indicated that doses ≥3 mg/kg suppressed lactation in rats so an assessment of the potential to adversely affect peri- and post-natal development in rats was carried out at doses of 0.1, 0.3 and 1.0 mg/kg/day. Maternal toxicity was seen at 0.3 and 1.0 mg/kg/day with slightly increased pup mortality at 0.3 and 1.0 mg/kg/day and lower mean pup weight from birth to day 35 post partum at 1.0 mg/kg/day. There were no apparent effect on the development and behavior of neither the offspring nor their reproductive ability.

- Other toxicity studies

2-Cl-AAI is a potential degradation product of the hydrochloride salt of rasagiline that tested positive in the bacterial reverse mutation assay when incubated in the presence of activated liver S9 fraction. To check for the potential presence of 2-Cl-AAI at the time of release and during stability studies of the drug product, a limit of 30 ppm was initially set as the release and stability specifications of the drug product. Further to a CHMP request to lower the specification limit of 2-Cl-AAI, the company decided to tighten the limit in the drug product to 20 ppm, which is well below the maximum daily value of 1.5μg being currently discussed as acceptable for genotoxic impurities.

Ecotoxicity/environmental risk assessment

The applicant presented a report according to which the crude Predicted Environmental Concentration of rasagiline mesilate in surface water is below 0.01 μg/l and therefore can be considered as an environmentally safe medicinal product.

Discussion on the non-clinical aspects

The precise mechanisms of action of rasagiline are unknown. In a series of in vitro and in vivo studies rasagiline was shown to be a highly potent, selective, irreversible monoamine oxidase type B (MAO-B) inhibitor. Rasagiline MAO-B inhibitory activity causes an increase in extracellular levels of dopamine in the striatum. The elevated dopamine level and subsequent increased dopaminergic activity are likely to mediate rasagiline's beneficial effects seen in models of dopaminergic motor dysfunction.

In addition, rasagiline showed some effects on cognition in animal models and neuroprotective effects in both in vitro and in vivo models. The neuroprotective activity of rasagiline is independent of its MAO inhibitory activity, and may be mediated by other mechanisms. Aminoindan, a major metabolite, which is not an MAO inhibitor, also showed an effect in models of impaired motor and cognitive functions. Nevertheless, animal models and *in vitro* data of neuroprotective effects have a very poor history of predicting clinical benefit.

Rasagiline is rapidly absorbed and quickly metabolized. It has a very large volume of distribution equivalent to several times total body water. Protein binding is in the region of 70-90% for animals and from 88-94% in man. The metabolic profile of rasagiline is qualitatively similar for animals and humans. The radioactive label of a ^{14}C-rasagiline is eliminated predominantly in the urine, 80-90% of the dose being recoverable.

After intravenous and oral dosing the principal manifestations of toxicity were related to the loss of selectivity for MAO-B (i.e. reduced food intake and weight gain and hyperactivity and/or aggression in rats).

In vitro genotoxicity studies were equivocal but studies with rasagiline mesylate or the combination of rasagiline with levodopa/carbidopa did not show genotoxic potential *in vivo*. The genotoxic potential of the combination was not evaluated in vitro.

Increased incidences of combined lung adenomas/adenocarcinomas were observed in mice. Incidence was within the range of historical control, exposure in animal was much more higher than in clinical situation and there were no signals in the rat study. Considering the equivocal genotoxic potential and the lack of plausible explanation for lung tumor development in mice, a further test addressing the involvement of a genotoxic mechanism in the lung tumorigenesis observed in mice has been requested.

To further investigate the possibility of a mouse-unique metabolite, that may be responsible for the results observed in the mutagenicity and carcinogenicity studies, the applicant initiated a study designed to compare rasagiline metabolites in mice and rats biofluids to those in humans. A minor metabolite, with a yet unknown structure was found in plasma and urine of mice. This metabolite at a much lower dose-relative levels was also found in rats, but was not found in human plasma and urine. Further studies to elucidate this metabolite's structure and then to evaluate its mutagenic potential are ongoing and will be submitted post-marketing.

Although the mechanistic explanation is still lacking, the fact that tumours were observed in mice only may reduce the concern regarding human safety. Genotoxic and carcinogenic data are summarised in the section 5.3 of the SmPC.

4. Clinical aspects

Introduction

Pharmacokinetic studies included:

- Pharmacokinetics of rasagiline (and aminoindan) in healthy subject following single dose (from 1 to 20 mg) and multiple doses (from 2 to 10 mg, for 10 days).
- Absorption, pharmacokinetics, plasma protein binding, metabolism and elimination of rasagiline following single doses of 3.2 mg ^{14}C-labeled rasagiline to healthy male volunteers.
- A bioequivalence study of the clinical trials and commercial formulation of rasagiline.
- A cross-over 'fed and fasting' study in healthy male volunteers.
- Multiple Dose Phase II Studies in Parkinson Patients (not Receiving Levodopa/Carbidopa Treatment, Receiving Chronic Levodopa/ Carbidopa Treatment, Receiving Chronic Levodopa Treatment, and Receiving Chronic Levodopa/Carbidopa or Levodopa/Benserazide Treatment)
- Population pharmacokinetic analyses in the target population were presented from two of the main clinical studies (TEMPO and PRESTO).
- Studies in renal impairment and hepatic impairment.
- Interaction studies with ciprofloxacin and theophylline.

Pharmacodynamic studies included:

- Evaluation of the MAO-B inhibitory properties within several PK studies,
- Study P94159 evaluated the potential for a pharmacodynamic interaction between rasagiline and tyramine in healthy volunteers.
- Study 132 was a ten week evaluation of the safety and tolerability of rasagiline 1 and 2 mg/day concomitantly administered with oral tyramine in 20 Parkinson's disease patients on levodopa treatment.

Pivotal clinical studies consisted of

- a on monotherapy study: TEMPO study,
- and 2 studies with rasagiline as add-on to levodopa – PRESTO and LARGO studies.

According with the documentation provided all studies were undertaken in accordance the principles of Good Clinical Practice.

Pharmacokinetics

The analytical method used capillary gas chromatography/negative ion chemical ionisation mass spectrometry. The lower limit of quantitation was 0.250 ng/mL and the assay was linear in the range 0.250 ng/nL to 25 ng/mL.

- Absorption – Bioavailability

Rasagiline is rapidly absorbed following oral administration, with an absolute bioavailability of approximately 36%, indicating that rasagiline undergoes a significant first-pass metabolism. Maximum plasma concentrations of rasagiline (C_{max} ~10 ng/mL) are reached at approximately 0.5 hours after oral administration (2 mg) in healthy subjects. In a mass balance study, after oral administration of 2 mg ^{14}C-rasagiline, at least 60% of the radioactive dose was absorbed.

The pharmacokinetics of 2 mg rasagiline administered following a high fat meal or under fasted conditions was compared. Administration of rasagiline with a high fat meal resulted in a decrease in extent of absorption (C_{max}) and systemic exposure (AUC) by about 60% and 20%, respectively. Considering that inhibition is irreversible and long-lasting and will increase over time in repeat dosing, the reduced C_{max} is not expected to affect the pharmacodynamic of the product. Considering that a decrease of 20% AUC should not have clinical consequence, no specific recommendation with regard to administration with food was considered necessary.

- Bioequivalence

A comparative *in vivo* bioequivalence study was performed demonstrating bioequivalence of the two 1 mg formulations: 1 mg Formulation II, used for the clinical trials and 1 mg intended for marketing tablet.

- Distribution

The pharmacokinetic disposition of rasagiline is best characterised by a two-compartment open model with first-order absorption and first-order elimination.

Rasagiline appears to be widely distributed, with a mean apparent volume of distribution (Vd) of about 243 l following a single dose IV administration (2 mg). The large Vd is related to the irreversible binding of rasagiline to MAO in the body and is consistent with the findings in the population PK studies.

The blood cell-to-plasma ratio for rasagiline derived radioactive material ranged from 0.1 to 1.2 with a mean ratio across time of 0.2 to 0.7, indicating that association and/or distribution of rasagiline and/or its metabolites into blood cells is not extensive.

- Metabolism and Elimination

Rasagiline is extensively metabolised in the liver following oral administration. *In vitro* studies have shown that CYP1A2 is the predominant P450 isoform involved in the metabolic elimination of rasagiline.

The primary human plasma metabolite formed following biotransformation of rasagiline is aminoindan. The proposed principal biotransformation pathways of rasagiline in human are N-dealkylation, hydroxylation of the indan ring, along with Phase II N or O-conjugation, including N-glucuronidation of the parent drug and of its metabolites.

There was no bioconversion of rasagiline mesylate (R enantiomer) to its S enantiomer within the human body, as determined in plasma samples for healthy volunteers dosed with rasagiline. Rasagiline is not metabolised to amphetamine or methamphetamine.

Rasagiline is eliminated with a half-life of about 0.6-2 hours and ranging from 0.3 to 3.5 hours across the 0.5 to 20 mg dose range examined following oral administration. Following a single IV dose (2 mg) in healthy subjects, the mean total body clearance (CL_{total}) for rasagiline is about 100 l/h (1668 ml/min). Less than 1% of the orally administered dose is excreted as unchanged drug in urine.

About 60% of the rasagiline derived radioactivity was excreted in urine and about 7% was excreted in the faeces over a 7-day period. Elimination was not complete in 7 days, and slow excretion, mostly faecal, continued with an estimated half-life of about 16 days. Prolonged excretion of radioactive material occurred possibly due to long-term irreversible binding of rasagiline to MAO enzyme in the body.

- Dose proportionality and time dependencies

Dose proportionality was assessed as part of the dose ranging studies in healthy subjects and PD patients. Rasagiline kinetics is apparently linear in the studied dose range of 0.5 to 2 mg per day.

- Special populations

In subjects with mild impairment of hepatic function (Child-Pugh class A), the apparent rasagiline clearance (CL/F) was reduced (42% decrease) resulting in increases in AUC (80%) and C_{max} (38%). In subjects with moderate impairment of hepatic function (Child-Pugh class B) the apparent rasagiline clearance was more markedly reduced (82% decrease) resulting in increases in AUC (568%) and C_{max} (83%), compared to subjects with no hepatic impairment. Since impaired hepatic function has major influence on the PK of rasagiline, rasagiline should be contraindicated in patient with severe impairement, be avoided in patients with moderate hepatic impairment and caution is necessary in patients with mild hepatic insufficiency.

In subjects with mild renal impairment (creatinine clearance [CLcr] = 50-80 mL/min) slightly higher AUC (38%) was observed, while C_{max} was unchanged. In subjects with moderate renal impairment (CLcr = 30-49 mL/min), AUC and C_{max} were 33% and 44% higher, respectively, compared to healthy subjects (CLcr >80 mL/min). In population PK studies serum creatinine clearance was tested as a covariate but found not to be significant. For the primary metabolite, aminoindan, similar and significant differences in PK were observed in both mild and moderate renal impaired groups. Since impaired renal function has little influence on the PK of rasagiline and aminoindan is not an MAO-B inhibitor, no change in dosage is required for mild and moderate renal impairment.

Due to rasagiline metabolism through CYP1A2, information regarding the potential interaction with smoking (CYP1A2 inducer) was performed in two population PK analyses performed as part of the pivotal studies PRESTO and TEMPO. In PRESTO analysis study, there was no significant effect of tobacco smoking on rasagiline pharmacokinetic. The results of the TEMPO analysis indicated that rasagiline clearance was increased in tobacco smokers by 39%. In both studies only 5% of the participants were smokers. The induction effect of tobacco on CYP 1A2 is therefore unclear. As a consequence, it should stated in the SPC that there is a risk that the plasma levels in smoking patients could be decreased, due to induction of the metabolising enzyme CYP1A2.

- Interaction studies

In vitro studies showed that rasagiline (1 µg/mL) caused no significant inhibition (<25% in all cases) of the model substrates, ethoxyresorufin, coumarin, tolbutamide, S-mephenytoin, bufuralol, chlorzoxazone, testosterone, and lauric acid. In addition, rasagiline did not cause mechanism-based inhibition of P450 isoforms. Therefore, rasagiline at therapeutic concentrations is unlikely to cause any clinically significant interference with substrates of CYP1A2, CYP2A6, CYP2C9, CYP2C19, CYP2D6, CYP2E1, CYP3A4, and CYP4A.

However, following co-administration of rasagiline and ciprofloxacin, a CYP1A2 inhibitor, the metabolism of rasagiline to aminoindan was inhibited. For rasagiline, this inhibition resulted in an increase in AUC of 83-98% (AUC0-inf to AUC0-t, respectively). Although C_{max} remained nearly unchanged, t_{max} was delayed by approximately 30 minutes (68%). For aminoindan, overall plasma levels were reduced, by 29% and 21% for C_{max} and AUC, respectively. As a general precaution, care must be taken when rasagiline is co-administered with compounds that inhibit CYP 1A2 because of the risk of increase of rasagiline plasma levels.

The lack of effect of rasagiline on metabolising enzymes was confirmed in a clinical study in which theophylline, a CYP1A2 substrate, was co-administered with rasagiline. There was no PK interaction between theophylline and rasagiline when the two drugs were administered concomitantly. Thus, no dose adjustment is necessary when theophylline and rasagiline are co-administered.

Rasagiline should not be administered along with other MAO inhibitors because of the risk of non-selective MAO inhibition that may lead to a hypertensive crisis.

Serious adverse events have been reported with the concomitant use of fluoxetine or fluvoxamine and MAO inhibitors including a selective MAO-B inhibitor. Therefore, in view of rasagiline's MAO inhibitory activity the concomitant administration of rasagiline and fluoxetine or fluvoxamine should be avoided.

Rasagiline has a high binding potential to human plasma proteins as indicated by an *in vitro* binding study that showed binding between 88 to 94% across the concentration range of 1-10,000 ng/ml. However, binding to isolated proteins, albumin and alpha-1-acid glycoprotein, which are the most common serum binding proteins for drugs, was low and ranged around 64% for albumin and 34-64% for alpha-1-acid glycoprotein. It is therefore unlikely that rasagiline will compete or will be displaced by any of the drugs that are known to be extensively bound to plasma proteins. *In vivo*, rasagiline is rapidly metabolized and the metabolites formed are of low binding activity. Moreover, rasagiline is administered at a very low dose of 1 mg/day, therefore, it should not have a potential for displacing other drugs in a clinically relevant manner.

Pharmacodynamics

- Mechanism of action

The precise mechanisms of action of rasagiline are unknown. Only the MAO-B inhibitory activity, which causes an increase in extracellular levels of dopamine in the striatum, was documented in human studies (see below). The other putative pharmacodynamic mechanisms are based on *in vitro* or animal data.

- Primary and Secondary pharmacology

In a series of *in vitro* and *in vivo* studies rasagiline was shown to be a highly potent, selective, irreversible monoamine oxidase type B (MAO-B) inhibitor.

The evaluation of the MAO-B inhibitory properties of rasagiline was done within the protocols of several pharmacokinetic studies. In healthy subjects and Parkinson's disease patients, rasagiline single or multiple administrations at all evaluated doses ranging from 0.5 mg/day to 10 mg/day significantly inhibits platelet MAO-B activity. MAO-B activity inhibition is maintained from at least one week to several weeks following cessation of rasagiline administration, with recovery between 2-6 weeks. The inhibition produced in PD patients taking or not levodopa is less marked than in healthy volunteers.

In Parkinson's disease patients on chronic levodopa treatment, multiple administration of rasagiline doses of 1 to 4 mg/day resulted in nearly complete MAO-B inhibition. In patients treated with 1 or 2 mg/day rasagiline, inhibition of MAO-B activity of >90% was reached within 1 to 3 weeks of dosing. No correlation of inhibition to dose was found in this study. Inhibition of platelet MAO-B activity was maintained for more than 1 month following dose cessation.

It is noteworthy that none of the putative effect of rasagiline, as anti-depressant and cognitive enhancer, suggested by the animal data, were specifically studied.

Pharmacodynamic interactions

Based on data with other MAO inhibitors, due to potential for pharmacodynamic interaction with other MAO inhibitors, sympathomimetic agents, antidepressants and the opioids pethidine and dextromethorphan, the following recommendation have been included in the SPC:

- Concomitant treatment with other monoamine oxydase inhibitors (MAOI) or pethidine is contraindicated,
- The concomitant use of rasagiline and fluoxetine or fluvoxamine should be avoided,
- The concomitant use of rasagiline and dextromethorphan or sympathomimetics including nasal and oral decongestants and cold remedies is not recommended,
- Antidepressants should be administered with caution.

The applicant provided data to support that 15-day washout is sufficient to eliminate the majority of the rasagiline effect on MAO; some of the effect might persist for longer but with unlikely clinical

relevance. Accordingly it is reasonable to propose a washout of 2 weeks before putative interacting products like SSRIs are initiated.

Tyramine interaction

The risk of hypertensive crisis with non-selective MAO inhibitors following intake of indirectly acting sympathomimetic amines such as tyramine is well recognized and can be potentiated in subjects receiving concomitant LD. Reversible MAOIs or doses of irreversible MAO-inhibitors that maintain selectivity for the B-form of the enzyme would not be expected to interact with tyramine.

As part of the rasagiline clinical program four clinical pharmacology studies were conducted to assess the potential pressor effect of tyramine when given concomitantly with rasagiline. One study was conducted in healthy volunteers and the others were conducted in PD patients (with and without levodopa); one pharmacodynamic interaction study (study 132), one sub-study of TEMPO and one sub-study of PRESTO. In all challenge studies a Holter monitor was used for continuous blood pressure measurements.

In the healthy volunteer study, the results showed that once daily 1 mg rasagiline did not increase the sensitivity for tyramine compared to placebo. An increase in sensitivity to tyramine was observed for subjects receiving 2 mg rasagiline and 10 mg selegiline and differences between the rasagiline and selegiline groups were not statistically significant.

Study 132 was conducted in 20 PD patients chronically treated with levodopa. Two patients on 2 mg in the study showed a tyramine pressor effect. Nevertheless, they were exposed to unrealistic conditions:

- at fasting (while administration of tyramine in food (particularly lipid) reduces by about three-fold the bioavailability of tyramine),
- with high doses of tyramine (75-225 mg compared to a maximum of 40 mg in a rich tyramine meal)
- and with double of the proposed clinical dose of rasagiline.

In the two tyramine challenge sub-studies of pivotal studies (monotherapy and adjunct to levodopa), a high dose tyramine challenge was performed following 6 months of treatment in order to show that MAO-B selectivity is maintained after long term exposure. The tyramine was given with a light meal to avoid extreme conditions. None of the patients had a tyramine pressor effect.

Finally, in the total non-tyramine restricted exposure to rasagiline (0.5 mg, 1 mg, 2 mg) database over 410 patient years on rasagiline alone and over 660 patient years on rasagiline and levodopa. None of these patients had event that could be considered a result of a potential tyramine/rasagiline interaction.

In conclusion, although tyramine/rasagiline interaction is not nil, it is sufficiently mild not to determine an unacceptable risk and rasagiline 1mg/day can be used without dietary tyramine restrictions.

Clinical efficacy

The primary objectives in the clinical development program were to show that rasagiline as monotherapy improves PD symptoms in early PD, and that it provides symptomatic benefit as adjunct therapy to LD in advanced PD complicated by motor fluctuations.

There were three main clinical studies:

- a monotherapy study: TEMPO study
- and 2 studies with rasagiline as add-on to levodopa – PRESTO and LARGO studies.

Study No.	Design	Treatment groups	Patients Entered/ Completed	Study Duration
TVP1012/232 (TEMPO Placebo-Controlled Phase)	Multi-center, double-blind, randomised, placebo-controlled/active-controlled, parallel group	*Placebo-Controlled Phase* Group 1: 1 mg qd for 26 weeks Group 2: 1 mg qd for 1 wk + 2 mg qd for 25 wks Group 3: Placebo for 26 weeks	134/125 132/124 138/133	56 weeks (Placebo-controlled for the first 26 weeks and double-blind active phase for the remaining 26 weeks)
TVP1012/232 (TEMPO Dose-Controlled Phase*)		*Dose-Controlled Phase** Group 1: 1 mg qd for 26 weeks Group 2: 2 mg qd for 26 weeks Group 3: 1 mg qd for 1 wk + 2 mg qd for 25 wks	124/120 124/118 132/122	
TVP1012/122 (LARGO)	Multi-center, double-blind, double-dummy, randomised, active- and placebo-controlled	Group 1: 1 mg qd for 18 weeks Group 2: Entacapone 200 mg with each levodopa dose for 18 weeks Group 3: Placebo for 18 weeks	231/208 227/197 229/194	18 weeks
TVP10121/133 (PRESTO)	Multi-center, double-blind, randomised, placebo-controlled, parallel group	Group 1: 0.5 mg qd for 26 weeks Group 2: 1 mg with qd for 26 weeks Group 3: Placebo for 26 weeks	164/142 149/132 159/140	26 weeks

*Patients who had received rasagiline during the placebo-controlled phase (groups 1 and 2) continued on the same treatment dose. Patients of group 3 who had originally been randomised to placebo began rasagiline treatment.

- Dose response study

No formal dose response studies were conducted. Based on experimental data on potency of rasagiline compared to selegiline and early clinical data in patients with doses up to 10 mg/day, the doses selected for the monotherapy, Phase III, double-blind, placebo-controlled TEMPO study were 1 and 2 mg/day. Since both rasagiline doses were found to be clinically comparable, the 1 mg/day was chosen as the recommended dose for monotherapy. Rasagiline 1 mg/day was chosen as the highest dose for the PRESTO and LARGO adjunct therapy studies in advanced PD patients experiencing LD-related motor fluctuations. A dose response evaluation of rasagiline 0.5 mg/day was included in the PRESTO study. 0.5 mg is significantly less efficacious than 1 mg.

- Main studies

Monotherapy – TEMPO study

METHODS

This was a North American, multicenter, randomized, double-blind, parallel, two-phase, Phase III clinical study. A 26-week, placebo-controlled treatment phase was followed by a 26-week dose-treatment phase (i.e. rasagi1ine 1 or 2 mg qd).

Objectives

The objectives of this study were to assess the efficacy, tolerability and safety of two doses of rasagiline in early PD patients who were not receiving or did not require levodopa/carbidopa (LD/CD) therapy.

Study Participants

To be included in the study, patients were required to have idiopathic PD with a severity of $\leq$3 in USA or <3 in Canada on the Modified Hoehn and Yahr scale. For at least six weeks prior to baseline, patients could not be treated with LD or dopamine agonists. Prior to baseline, washout periods from amantadine, LD or dopamine agonists, selegiline, selective serotonin re-uptake inhibitors (SSRI), other antidepressants (except for amitriptyline and trazodone) and meperidine (pethidine), were required.

Exclusion Criteria included: Anti-PD medications except for anti-cholinergics. Clinically significant malignancy, vascular disorders, psychiatric illness and abnormal clinically significant laboratory test results.

Treatments

Early stage PD patients were initially randomized in a 1:1:1 ratio to one of two daily doses (1 or 2 mg) of rasagiline or placebo. In the second phase, patients who had originally been randomized to placebo began treatment with a dose of 1 mg/day for one week and then were treated with 2 mg/day.

Outcomes/endpoints

The primary efficacy end-point was the change in total Unified Parkinson's Disease Rating Scale (UPDRS) from baseline to termination visit (Week 26). UPDRS has a total of 44 items in three categories, mental function, daily living, and neurological (motor) examination; each item is scored 0 – 4, giving a possible range of 0 – 176, with higher scores denoting worse disease.

Secondary criteria were responder analysis (a posteriori definition), change in the UPDRS sub-scales, time to the need for levodopa therapy, and the proportion of patients requiring levodopa.

Regarding the dose-controlled phase and overall study period, the efficacy measure was the change in total UPDRS scores, calculated from baseline (Week 0) to last observed value before additional anti-PD therapy was administered, comparing rasagiline 1 and 2 mg/day treatment groups with placebo/2 mg group.

Sample size

The sample size was based on the assumption that there would be a treatment advantage of three or more points in the UPDRS score of rasagiline 2 mg, with an advantage of 0 – 3 for 1 mg and a standard deviation of 8.75 UPDRS scores on the basis of published studies. Given α set at 0.05 and approximately 80% power then 120 patients were needed per treatment arm.

Randomisation

Eligible patients were allocated to one of the three treatment groups using a randomization scheme with blocks stratified by center.

Statistical methods

Intent-To-Treat cohort (ITT) includes all patients who have been randomized.

Completers cohort (CO) includes all the patients who completed 26 weeks of the placebo-controlled phase and patients that needed LD therapy during that phase and entered the dose-treatment phase.

Per Protocol Cohort (PP) includes all patients who completed 26 weeks of the placebo-controlled phase (including patients that needed LD therapy) and did not have major protocol violations.

For the ITT cohort, the LOCF approach was applied to account for missing data at the placebo-controlled phase termination (Week 26). For the Dose-Treatment phase and Efficacy Cohorts, the LOCF approach was applied to account for missing data at study termination (Week 52).

RESULTS

The baseline characteristics are shown in the table below.

	Rasagiline 1 mg	Rasagiline 2 mg	Placebo
No. Recruited	134	132	138
No. completed (6-month)	111	105	102
Age (SD:) in years	61.6 (10.3)	60.4 (11.4)	60.5 (10.8)
Proportion male (%)	67	56	67
Disease duration	0.93 (1.2)	1.16 (1.3)	0.94 (1.1)
Baseline total UPDRS	24.69 (11.25)	25.89 (9.54)	24.54 (11.61)

Participant flow

Four hundred and seventy-three (473) patients were screened. Of these, 404 (84%) patients enrolled into this study.

Around 80 % of patients completed the 6-month, placebo-controlled phase. The differences between treatment groups in the number of patients with premature termination or the time on study to termination were not statistically significant.

Recruitment

Despite the relatively wide entry criteria the actual patients being recruited were mildly affected as evidenced by the mean UPDRS motor score of about 25. Around 1/3 of all enrolled patients had received dopaminergic products before enrollment and 1/6 of enrolled patients were in stage Hoehn and Yahr III (presence of abnormal postural reflexes) that is an indicator of at least moderate disease.

Baseline data

The mean age was 61 years (range 32-92); 64% males. About 95% of the patients were Caucasians.

Disease duration for all treatment groups was similar. On average, mean disease duration in all treatment groups was one year at study entry.

Baseline disease characteristics were comparable between treatment groups. No statistical significant differences were demonstrated between groups, except for higher scores in the rasagiline 2 mg group of UPDRS mental scale and a boundary significance of Severity of Illness scale.

Outcomes and estimation

The main efficacy results are shown in the table below.

	Rasagiline 1 mg	Rasagiline 2 mg	Placebo
No. Recruited	134	132	138
Adjusted mean change from baseline in total UPDRS [95% CI]	-0.13 [-1.16, 0.91]	0.51 [-0.55, 1.57]	4.07 [3.04, 5.1]
Adjusted mean change in UPDRS Motor	-038	0.65	2.33
Adjusted mean change in UPDRS ADL	0.16	-0.02	1.20

The principal statistical analysis compared the mean change from baseline in total UPDRS for each of the active-treatment groups to placebo (two contrasts) using ANCOVA adjusted for baseline UPDRS, treatment, center and treatment-by-center interaction. Following 26 weeks of treatment, the change from baseline UPDRS differed significantly between the treatment group and the placebo ($p<0.0001$ for both contrasts using Hochberg's Step-up Bonferroni procedure for multiple comparisons). The adjusted mean changes from baseline in total UPDRS score are presented in the table above. The treatment effect exerted by 1 and 2 mg rasagiline was therefore -4.20 (95% CI:[-5.66,-2.73]) and -3.56 (95% CI: [-5.04,-2.08]), respectively.

Responder Analysis: Non-responders were defined as patients with a worsening of 3 or more points in total UPDRS. All other patients were considered responders. About two-thirds of rasagiline-treated patients but only approximately half of the placebo-treated patients were classified as responders at the end of the 26-week, placebo-controlled phase. The difference between each one of the rasagiline groups and placebo was statistically significant (1 mg vs. placebo: Odds Ratio 2.2, 95% CI [1.3, 3.7] p=0.0038; 2 mg *vs.* placebo: Odds Ratio 2.5, 95% CI [1.4, 4.2] p=0.0011 using Hochberg's Step-up Bonferroni procedure for multiple comparisons.

Most results on secondary end-points are consistent with the primary-ones favoring the superiority of rasagiline over placebo.

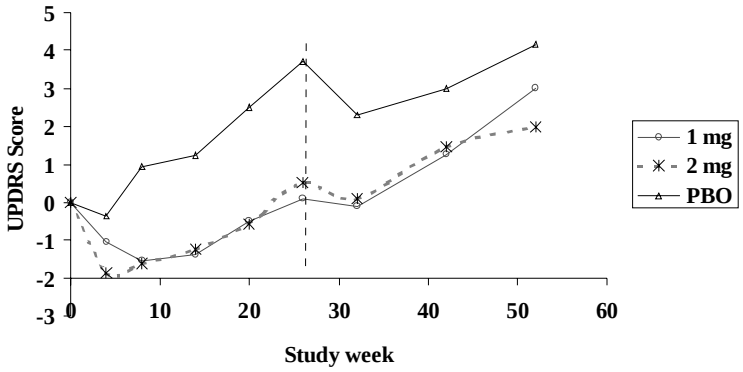
- Following the 26-week placebo-controlled phase, mean UPDRS Motor score was not significantly different from baseline in patients on both rasagiline doses. UPDRS Motor scores were increased by a mean of 14% in placebo-treated patients. Compared to placebo, the adjusted mean treatment effect was (-2.71, 95% CI [-3.87, -1.55] $p<0.0001$) in patients treated with 1 mg/day and (-1.68, 95% CI [-2.85, -0.51] p=0.0050) in patients treated with 2-mg/day rasagiline.
- Following 26-week treatment period, patients on both doses of rasagiline maintained mean ADL scores similar to baseline, while patients on placebo experienced an increase of about 19% in their mean ADL score. Compared to placebo, significant treatment effects were detected in the 1 mg/day (-1.04, 95% CI [-1.60, -0.48], p=0.0003) and the 2 mg/day (-1.22, 95% CI [-1.79, -0.65] $p<0.0001$) treatment groups.
- In the Global Improvement evaluation, following 26 weeks of treatment, a significantly ($p = 0.015$) greater percentage of patients (72.9%) treated with 1 mg rasagiline experienced global improvement or no decline compared to placebo (62.3%).
- Following 26 weeks of treatment, Quality of Life (QOL) scores did not differ significantly from baseline in actively treated patients; a 9.6% increase from baseline in QOL score was observed in patients treated with placebo indicating deterioration Compared to placebo, a significant treatment effect was observed in patients treated with 1 mg /day (-2.91, 95% CI [-5.19, -0.64], p=0.0122) and 2 mg /day (-2.74, 95% CI [-5.02, -0.45], p=0.0191) rasagiline.

No difference between groups was detected regarding the time to LD need. The proportion of patients who did not need LD was similar in treatment groups (88.8% in the 1 mg group and 83.3% in each of the other groups).

Dose controlled phase:

382 patients completed the placebo controlled phase of the study, of which 380 continued into the active treatment phase. The figure below shows mean change from baseline in total UPDRS.

Study 232 mean change from baseline in Total UPDRS Efficacy cohort
scheduled visit LOCF (vertical line shows change from placebo control)



Long-term open extension

The long-term open extension of the TEMPO study enabled the follow-up of patients for more than 5 years. Of the patients who completed 2 years of treatment 81.5% were still on rasagiline, without levodopa treatment. Of the patients who completed 5 years of treatment 40% were still on rasagiline, without levodopa treatment.

Adjunct treatment trials

PRESTO study

METHODS

PRESTO was a multicenter, US and Canada, double-blind, randomized, placebo-controlled, parallel-group study, for the efficacy, tolerability and safety of rasagiline mesylate in levodopa treated Parkinson's disease patients with motor fluctuations.

Objectives

To evaluate the efficacy, tolerability, and safety of 0.5 mg and 1 mg of rasagiline, once daily, versus placebo in subjects with levodopa-treated Parkinson's disease (PD) and motor fluctuations.

Study Participants

Eligible subjects were aged 30 years or older and had idiopathic PD confirmed by the presence of at least two of the cardinal signs for PD (resting tremor, bradykinesia and rigidity) graded on the Modified Hoehn and Yahr as stage <5 in the "OFF" state. Included were subjects who chronically used levodopa and experienced motor fluctuations averaging at least 2.5 hours daily in the "OFF" state, confirmed by the baseline home diaries. Subjects were required to demonstrate competence in diary completion. Subjects were permitted to maintain stable doses of other anti-PD medications. Prior to baseline, subjects were required to discontinue and washout of selegiline, antidepressants meperidine (pethidine), tolcapone, sympathomimetics, dextromethorphan, St. John's Wort and

gentamicin. Amitriptyline, trazodone, citalopram, paroxetine, and sertraline were allowed at stable, low doses.

Any of the following excluded subjects from participation in the study: severe disabling dyskinesias, previous use of rasagiline, neurosurgical intervention for PD within the 12 months preceding the baseline visit, clinically significant malignancy, severe hypertension, severe orthostatic hypotension vascular disorders, psychiatric illness (Beck depression scale $\geq$ 15), cognitive impairment (MMSE score $\leq$ 24) and clinically significant abnormal laboratory test results.

Treatments

Following a screening visit and screening period to ensure that subjects met all enrollment criteria and could accurately complete home diaries, subjects were randomly assigned to treatment at the baseline visit. The levodopa dose could be decreased for the first 6 weeks of the study period at the discretion of the investigator, but remained constant for the last 20 weeks.

Outcomes/endpoints

The primary efficacy criterion was change from baseline in OFF time as recorded by use of home diaries filled in for half hourly intervals for the three days prior to clinic visits at baseline, at weeks 6, 14 and 26 (termination).

Secondary criteria were Global Improvement evaluated by the examiner; change from baseline in UPDRS ADL score in the OFF state; change from baseline UPDRS Motor score in the ON state; change from baseline in PD-QUALIF (a quality of life scale).

Sample size

The power calculations for the study assumed that the pooled standard deviation in OFF time would be two hours with α set at 0.5 and β at approximately 80% then 150 patients per treatment arm would be capable of detecting a difference of 45 minutes in OFF time between treatment and placebo. Inference testing was by Student's t-test using the Bonferroni modification.

Blinding (masking)

A tyramine tolerance sub-study was performed in 55 subjects on the last day of the 26-week study period. To prevent unblinding in case of a tyramine reaction, the investigator and coordinator roles for this sub-study were not carried out by the same individuals who performed these roles in the PRESTO study.

Statistical methods

The principal statistical analysis of the primary endpoint was an Analysis of Covariance (ANCOVA) adjusting for baseline mean total daily "OFF"time. The adjusted mean of the changes observed in each of the rasagiline dose groups (two contrasts) were compared to placebo by applying an ANCOVA model on the primary endpoint as dependent variable. The model included the following fixed effects: treatment group, center and baseline mean total daily "OFF" time. The primary efficacy endpoint was analyzed for the 3 types of cohorts, the Intent-To-Treat cohort (the principal cohort for inference), the Completers cohort and the Per-Protocol Cohort. Statistical tests were performed at the 5% significance level (two-tailed). The Hochberg's Step-up modification to Bonferroni method was used to protect from inflation in the type I error resulting form multiple contrasts testing.

Secondary endpoints were analyzed using ANCOVA. For the Responders Analysis, a Logistic Regression was performed.

Except for the primary efficacy analysis (for which the mean of all measurements available during treatment was calculated) the Last Observation Carried Forward (LOCF) approach was used to account for missing interim data, for subjects with at least one post-randomization evaluation. Subjects without post randomization efficacy endpoint evaluation were excluded from the analyses of this endpoint.

RESULTS

Participant flow

A total of 606 subjects were screened. Of these, 472 were randomized to one of two once-daily doses of rasagiline (0.5 mg and 1 mg) or placebo; 164 (35%) received 0.5-mg/day rasagiline, 149 (31%) received 1 mg/day rasagiline and 159 (33%) received placebo.

From the 0.5 mg/day rasagiline treatment group 142 subjects (87%) completed the full duration of the study and 22 subjects prematurely withdrew from the study. From the 1 mg/day rasagiline treatment group 132 subjects (89%) completed the study and 17 subjects prematurely withdrew from the study. From the placebo treatment group 140 subjects (88%) completed the study and 19 subjects prematurely withdrew from the study.

The most common reason for prematurely withdrawing from the study was the experiencing of AEs with an overall incidence of 7%. The 0.5 mg/day rasagiline treatment group had the largest withdrawal due to AEs (9%). This was followed by the 1 mg/day rasagiline treatment group (6%) and then by the placebo treatment group (5%). Premature withdrawal due to the worsening of PD symptoms occurred with the highest incidence in the placebo treatment group (4%) compared to an incidence of approximately 1% in each of the rasagiline treatment groups. The withdrawal of subject consent occurred with an incidence of approximately 2% in each treatment group.

Baseline data

There were no statistically significant differences between the treatment groups regarding demographic and clinical variables at baseline including age, sex, PD duration, levodopa treatment duration and levodopa total daily dose, fluctuation and dyskinesia duration, total daily "ON" and "OFF" time and UPDRS scores.

The great majority (above 90%) of subjects were Caucasians. 65 % were male patients. Mean age ranged from 63 to 65 years. Mean PD duration was of approximately 9 years. The study population received approximately a total daily levodopa dose of 794 mg and spent a mean 6.1 hours of waking hours in the "OFF" state.

Outcomes and estimation

Efficacy results are summarised in the two following tables providing treatment effect of rasagiline; 0.5 mg and 1 mg over placebo.

Rasagiline 0.5 mg over Placebo

Endpoint Type and Definition			**Treatment Effect**	**p-value**	**95% CI**
Primary endpoint: Change from Baseline to Treatment in Mean Total Daily "OFF" Time		ITT Cohort	-0.49 hours	p = 0.0199	-0.91, -0.08
		PP Cohort	-0.62 hours	p = 0.0087	-1.08, -0.16
		CO Cohort	-0.54 hours	p = 0.0156	-0.97, -0.10
Secondary Efficacy Endpoints	Global Improvement by Examiner at Termination		-0.39 units	p = 0.0027	-0.64, -0.13
	Change from Baseline to Termination in UPDRS ADL During "OFF" State		-1.20 units	p = 0.0075	-2.08, -0.32
	Change From Baseline to Termination in UPDRS Motor During "ON" State		-2.91 units	p = 0.0007	-4.59, -1.23
	Change from Baseline to Termination in QOL (PD-QUALIF)		-2.18 units	p = 0.0651	-4.49, 0.14

Rasagiline 1 mg over Placebo

Endpoint Type and Definition			Treatment Effect	p-value	95% CI
Primary endpoint: Change from Baseline to Treatment in Mean Total Daily "OFF" Time		ITT Cohort	-0.94 hours	p < 0.0001	-1.36, -0.51
		PP Cohort	-1.08 hours	p < 0.0001	-1.55, -0.61
		CO Cohort	-0.94 hours	p < 0.0001	-1.38, -0.49
Secondary Efficacy Endpoints	Global Improvement by Examiner at Termination		-0.68 units	p < 0.0001	-0.94, -0.42
	Change from Baseline to Termination in UPDRS ADL During "OFF" State		-1.34 units	p = 0.0040	-2.24, -0.43
	Change From Baseline to Termination in UPDRS Motor During "ON" State		-2.87 units	p = 0.0011	-4.58, -1.16
	Change from Baseline to Termination in QOL (PD-QUALIF)		-1.48 units	p = 0.2229	-3.86, 0.90

The difference between rasagiline and placebo treatments (in favor of rasagiline) in the change from baseline in the total daily "OFF" time is 0.94 hours for the 1 mg/day rasagiline treatment group and 0.49 hours for the 0.5 mg/day rasagiline treatment group. The beneficial effect of rasagiline is present across all study cohorts (ITT, Per Protocol and Completers) representing the internal consistency of the data.

The superiority effect detected in the 1 mg/day rasagiline-treatment arm compared to the 0.5 mg/day rasagiline-treatment arm suggests a dose relationship.

The beneficial effect of 1 mg/day rasagiline over placebo was already present at week 6, the first post-randomisation diary visit and was maintained across all study visits including the termination visit. The beneficial effect is obtained even though all the subjects were optimized on their levodopa treatment and most subjects were taking additional antiparkinsonian medications including dopamine agonists and/or entacapone, which themselves have the ability to improve fluctuations.

The clinical relevance of the primary endpoint data is confirmed by the responder analysis, based on the percentages of subjects with an improvement in total daily OFF time of at least 60 minutes. 65% of subjects from the rasagiline 1 mg/day treatment group, 59% of subjects from the 0.5 mg/day rasagiline treatment group, and 45% of subjects from the placebo treated arm were responders. Logistic Regression Analysis resulted in statistically significant odds ratios. For the 1 mg rasagiline treatment group an odds ratio of 2.5 in favor of rasagiline over placebo (p=0.0005, 95% CI: 1.49, 4.23) was obtained. For the 0.5 mg/day rasagiline treatment group, the odds of being a responder are 1.9 times higher (p=0.0110, 95% CI: 1.16, 3.14) compared with placebo treatment.

The reduction in the total daily "OFF" time corresponds closely to the increase in the total daily "ON", and the increase in the total daily "ON" time is due primarily to an increase in "ON1" time i.e., "ON" without dyskinesia or without troublesome dyskinesia.

Analyses of the secondary endpoints that were adjusted for multiplicity have demonstrated an overall statistically significant treatment effect that was attributable to rasagiline treatment for all 3 clinical secondary endpoints (Global Improvement rated by the Examiner, UPDRS ADL in "OFF" state, and UPDRS Motor in the "ON" state), although there is no evidence confirming a treatment effect for the 4th secondary endpoint measuring subjects quality of life with the PD-QUALIF scale.

LARGO study

METHODS

LARGO is a multicenter (Europe, Israel and Argentina), double-blind, double dummy, randomized, placebo and entacapone-controlled, 3 parallel groups study, for the efficacy, tolerability and safety of rasagiline mesylate in levodopa-treated Parkinson's disease patients with motor fluctuations.

Objectives

To evaluate the efficacy, tolerability, and safety of rasagiline mesylate versus placebo in subjects with levodopa-treated Parkinson's disease (PD) and motor fluctuations. The comparison between entacapone to placebo serves for validation and exploratory purposes only.

Study Participants

Male and female subjects, aged 30 years or older, with idiopathic PD, taking optimized levodopa/dopa decarboxylase inhibitor therapy, who experienced motor fluctuations averaging at least 1 hour daily in the "OFF" state during waking hours (not including morning akinesia) and graded on the Modified Hoehn and Yahr as stage <5 in the "OFF" state, were eligible. Included were subjects who chronically used levodopa together with a peripheral decarboxylase inhibitor and experienced LD-related motor fluctuations for at least 1 hour daily in the "OFF" state during waking hours (not including morning akinesia), confirmed by the baseline 24-hour diaries. Subjects were required to demonstrate competence in diary completion. Subjects who presently or previously used entacapone were not eligible for the study. Subjects were permitted to maintain stable doses of other anti-PD medications. Subjects were required to undergo washout prior to baseline from selegiline, antidepressants (except amitriptyline, and trazodone at low doses), meperidine (pethidine), tolcapone, sympathomimetics, dextromethorphan, St. John's Wort and gentamicin.

Treatments

Following a screening visit subjects started on a placebo run-in period of at least 2 weeks during which they were administered a placebo for rasagiline once daily before breakfast, as well as a placebo for entacapone with each levodopa dose. Eligible subjects were then randomized to the rasagiline, entacapone or placebo treatment groups for 18 weeks of double-blind treatment.

Levodopa dosage could be decreased during the first 6 weeks of the study period at the discretion of the investigator, but had to remain constant for the last 12 weeks.

Outcomes/endpoints

Subjects attended study visits at 3, 6, 10, 14 and 18 weeks after baseline for efficacy and safety monitoring. A home 24-hour diary in which subjects rated themselves as "ON without dyskinesias or without troublesome dyskinesias", "ON with troublesome dyskinesias", "OFF", or "asleep" every half hour was completed for 3 consecutive days immediately prior to randomization (baseline), and weeks 6, 10, 14 and 18.

The primary efficacy endpoint was the change from baseline through the treatment period in the mean total daily "OFF" time, as measured by 24-hour diaries.

Secondary Outcome Measures included Global Improvement by Examiner, Change from baseline to termination in UPDRS ADL during "OFF" phases and Change from baseline to termination in UPDRS Motor during "ON" phases.

There were additional efficacy outcome measures including a responder analysis, and, safety and tolerability measures.

Sample size

The same assumptions as for PRESTO was planned in the clinical trial protocol, but, an assessment of the variance magnitude was performed after one third of the subjects had completed 18 weeks of treatment. This assessment revealed that, in order to preserve the original power of the study and to avoid the continuation of a futile study, the design should be upsized to enroll a total of 700 patients, 250 more than originally planned. Therefore, a total of 687 subjects were actually randomized into this study.

Statistical methods

The statistical analysis of the primary endpoint was performed on the intent-to-treat (ITT), per-protocol (PP) and complete (CO) cohorts using an analysis of covariance (ANCOVA) accounting for baseline mean total daily "OFF" time. The adjusted means of the changes observed in the rasagiline mesylate 1 mg treatment group was compared with placebo by performing a single degree of freedom comparison in a model including the three treatment groups. Statistical tests were performed at the 5% (two-tailed) significance level.

To avoid inflation of Type-1 error due to multiple endpoints testing, the analyses of secondary endpoints were performed according to a hierarchical order, with the testing of each endpoint only being conducted if the prior endpoint demonstrated a statistically significantly beneficial effect of rasagiline compared with placebo. Secondary endpoints were analyzed using ANCOVA except for the Global Improvement by Examiner for which an ANOVA was performed.

Except for diary derived endpoints for which the mean of all measurements available during treatment was calculated, the Last Observation Carried Forward (LOCF) approach was used to account for missing data at week 18 for subjects with at least one post-randomization evaluation. Subjects without post-randomization efficacy endpoint evaluation were excluded from the analyses of this endpoint.

RESULTS

Participant flow

A total of 687 subjects were randomised: 231 subjects entered the rasagiline treatment group, 227 subjects entered the entacapone treatment group, and 229 subjects entered the placebo treatment group.

From the rasagiline treatment group 208 subjects (90%) completed the full duration of the study and 23 subjects prematurely withdrew from the study. From the entacapone treatment group 197 subjects (87%) completed the study and 30 subjects (13%) prematurely withdrew from the study. From the placebo treatment group 194 subjects (85%) completed the study and 35 (15%) subjects prematurely withdrew from the study.

The most common reasons for prematurely withdrawing from the study were subject withdrawal of consent and the experiencing of AEs, each with an overall incidence of 4.9%. The entacapone treatment group had the largest withdrawal due to AEs (7.0%). This was followed by the placebo treatment group (4.8%) and then by the rasagiline treatment group (3.0%). The placebo treatment group had the largest withdrawal of subject consent (6.6%). This was followed by the rasagiline treatment group (5.2%) and then by the entacapone treatment group (3.1%).

Baseline data

There were no statistically significant differences between the treatment groups regarding demographic and clinical variables at baseline including age, sex, PD duration, levodopa treatment duration and levodopa total daily dose, fluctuation and dyskinesia duration, total Daily "ON" and "OFF" times and UPDRS scores.

The great majority (above 90%) of subjects were Caucasians. 62 % were male patients. Mean age ranged from 63 to 65 years. Mean PD duration was of approximately 9 years. The study population received approximately a total daily levodopa dose of 707 mg and spent a mean 5.6 hours of waking hours in the "OFF" state.

Outcomes and estimation

Efficacy results over placebo are summarised in the following table.

Rasagiline 1 mg over Placebo

Endpoint Type and Definition			Treatment Effect	p-value	95% CI
Primary endpoint: Change from Baseline to Treatment in Mean Total Daily "OFF" Time		ITT Cohort	-0.78 hours	p = 0.0001	-1.18 to-0.39
		PP Cohort	-0.71 hours	p = 0.001	-1.14 to -0.29
		CO Cohort	-0.72 hours	p = 0.0006	-1.12 to -0.31
Secondary Efficacy Endpoints	Global Improvement by Examiner at Termination		-0.49 units	p< 0.0001	-0.68 to -0.31
	Change from Baseline to Termination in UPDRS ADL During "OFF" State		-1.71 units	p < 0.0001	-2.49 to -0.93
	Change From Baseline to Termination in UPDRS Motor During "ON" State		-2.94 units	p < 0.0001	-4.28 to -1.60

The results of this trial demonstrate that daily treatment with 1 mg of rasagiline reduces the total daily "OFF" duration by a mean 0.78 hours in levodopa-treated Parkinson's diseased patients with motor fluctuations. This clinically beneficial effect of rasagiline is present across all study cohorts representing the internal consistency of the data.

This study has also demonstrated the statistically significant beneficial effect (reduction of 0.80 hours for entacapone over placebo ($p < 0.0001$, 95% CI:-1.20 to -0.41) of the active study comparator, entacapone 200 mg, administered orally with each levodopa dose, providing additional evidence of the adequacy of the conduct of this clinical trial.

The beneficial effect of rasagiline over placebo was already seen at week 6, the first post-randomization diary visit, and persisted across all study visits including the termination visit.

The clinical relevance of the primary endpoint data is confirmed by the exploratory responder analysis, based on the percentages of subjects with an improvement in total daily "OFF" time of at least 60 minutes. 51% of subjects from the rasagiline treatment group, 45% of subjects from the entacapone treatment group and 32% of subjects from the placebo treatment group were responders. An odds ratio of 2.5 in favor of rasagiline over placebo ($p < 0.0001$, 95% CI: 1.62 to 3.85) was obtained.

The reduction in the "OFF" time is mirrored by the increase in the total daily "ON" and "ON1" times and is not accompanied by an increase in unwanted troublesome dyskinesia. Analyses of the secondary endpoints that were adjusted for multiplicity have demonstrated an overall statistically significant treatment effect that can be attributed to rasagiline treatment for all 3 secondary endpoints: Global Improvement by the Examiner, UPDRS ADL in "OFF" state, and UPDRS Motor in the "ON" state.

Rasagiline treatment has also shown a slight but statistically significant decrease in the mean total daily levodopa dose. A similar decrease is seen for entacapone treatment. Larger reductions in the daily levodopa dose following entacapone treatment have been seen in previous studies in which reductions were permitted according to the design of the study. This study did not permit the reduction except in the case of intolerance during the first 6 weeks of the study.

- Clinical studies in special populations

Means total daily "OFF" time at baseline, during treatment and change from baseline were compared according to the cut-off age below 70-years old and above that age in PRESTO and LARGO studies. The subgroup analyses by age suggests that rasagiline is equally efficacious on those above and below 70 years old.

- Discussion on clinical efficacy

Based on the known risk of hypertensive crisis with non-selective MAO inhibitors following intake of indirectly acting sympathomimetic amines such as tyramine, the risk of rasagiline interaction with tyramine was investigated. An increase in sensitivity to tyramine was observed in two healthy volunteers in a pharmacodynamic study. Nevertheless, the applicant has shown that two episodes of tyramine pressor response on 2 mg rasagiline occurred in unrealistic conditions which are unlikely under every day circumstances. In addition, in the total non-tyramine restricted exposure to rasagiline in the clinical programme, which is over 660 patient years on rasagiline and levodopa and over 410 patient years on rasagiline alone. None of these patients had any event that could be considered a result of a potential tyramine/rasagiline interaction. In conclusion, although tyramine/rasagiline interaction is not nil, rasagiline 1mg/day can be used without dietary tyramine restrictions.

The clinical development of rasagiline includes 3 pivotal trials; one in monotherapy in early PD patients and 2 as add-on therapy to levodopa in fluctuators. The 3 trials together demonstrate that rasagiline has a consistent anti-parkinson effect.

The 2 pivotal trials where rasagiline was tested as add-on to levodopa demonstrates that rasagiline reduces OFF time (1h in more than 50% of patients) and increases ON time without dyskinesias more that it increases ON with dyskinesias. The effect size of rasagiline 1 mg/d is similar to that obtained with entacapone. The mean difference compared to placebo, between 0.78 h and almost 1 h, is considered clinically relevant. The responder analysis supports the clinical relevance: improvement of at least 1.5 h in about 40% of the patients both for rasagiline and entacapone and improvement of at least 2h in 30% of actively treated. Efficacy in the treatment of motor fluctuations is therefore supported by the PRESTO and LARGO studies. Nevertheless, there was no controlled trial targeting the population of non-fluctuators that need further treatment of Parkinsonism signs, nor a controlled trial targeting patients with non-dose dependent fluctuations. Therefore, the therapeutic indication should be restricted to patients with motor fluctuations of the wearing-off type.

The monotherapy trial, which includes a 6-month placebo controlled phase and a further 6-month double-blind phase without placebo arm, shows an advantage over placebo for rasagiline at 6 months that is maintained at 1 year. This advantage is about 4 points in the total UPDRS (primary end-point) and 2.5 points in UPDRS motor, as compared to placebo. The clinical relevance of effect was questioned, considering that the size of the effect is modest (e.g. compared to historical data of other medicinal products, in particular dopamine agonists) and there was no active comparator arm in the TEMPO study.

The absence of a 3 arm trial including an active comparator in the monotherapy study has been justified by the applicant by the fact that the CHMP scientific advice for rasagiline (November 1999) and the CHMP Note for Guidance on Clinical Investigation of Medicinal Products in the Treatment of PD (adopted in December 1998 for coming into operation in June 1999) came too late to influence the design of the TEMPO study which enrolment started in November 1997 and ended in November 1999. Furthermore, particularly when the study started, there was no consensus on an appropriate comparator.

It was acknowledged that the effect of rasagiline in monotherapy seems smaller than the effect of LD or dopamine agonists. On the other hand rasagiline effect seems at least comparable to selegiline effect based on bibliographical data and selegiline is a MAO-B inhibitor widely recognised in the treatment of early PD. In any case, without direct comparison, no formal conclusion can be drawn with regard to rasagiline efficacy compared to other products. It was also noted that the placebo deterioration rate is in line with the natural history of the disease (around 10 UPDRS points / year). The clinical relevance of the effects observed on total UPDRS and the UPDRS motor are supported by consistent effects seen in the responder analysis and on UPDRS ADL, as well as with a quality of life scale (PDQUALIF - Welch, 2003) where statistically significant treatment effect in favor of rasagiline (a reduction of 2.92 units, $p = 0.0122$, 95% CI: 0.64, 5.19) was obtained. The lack of effect at 6 months for delaying LD use may be explained by the fact that it was a too short timeframe to see an effect on this end-point. The extension phase is supportive of a sparing effect because 69% of patients are still on monotherapy at one year, and 46% at 2 years.

Clinical safety

- Patient exposure

A total of 1864 subjects who participated in the clinical development program of rasagiline were included in the safety database. Of these, 1453 were exposed to rasagiline for 1583.7 subject years. A total of 596 subjects received placebo for 220.6 subject years and 227 subjects received entacapone for a total of 153.2 subject years. In the double blind placebo controlled studies 529 patients were treated with rasagiline 1 mg/day for 212 patient years.

- Adverse events

Monotherapy

Adverse reactions reported with a higher incidence in placebo-controlled studies, in patients receiving 1 mg/day rasagiline as monotherapy are presented in the table below with the adverse reaction incidence (% of patients) in rasagiline vs. placebo, respectively.

AEs in patients not on levodopa

	Adverse event	Rasagiline 1 mg (n=149) %	Placebo (n = 151) %
Body as a whole	Headache	14.1	11.9
	Flu syndrome	6.0	0.7
	Fever	2.7	1.3
	Malaise	2.0	-
	Neck pain	2.0	-
	Allergic reaction	1.3	0.7
Cardiovascular system	Angina pectoris	1.3	-
	Cerebrovascular accident	0.7	-
	Myocardial infarct	0.7	-
Digestive system	Dyspepsia	6.7	4.0
	Anorexia	1.3	-
Hemic and lymphatic system	Leucopaenia	1.3	0
Musculoskeletal system	Arthralgia	7.4	4.0
	Arthritis	2.0	0.7
Nervous system	Depression	5.4	2.0
	Vertigo	2.0	0.7
Respiratory system	Rhinitis	2.7	1.3
Special senses	Conjunctivitis	2.7	0.7
Skin and appendages	Contact dermatitis	1.3	-
	Vesiculabullous rash	1.3	-
	Skin carcinoma	1.3	0.7
Urogenital system	Urinary urgency	1.3	0

Reflecting the increased dopaminergic effect of the higher rasagiline dose, the most common AEs in the 2 mg group included abnormal dreams (<1% in 1 mg group vs. 3% in 2 mg group), vomiting (1% for 1 mg vs. 3% for 2 mg) and sleep disorder (4% in 1 mg group vs. 6% in 2 mg group), somnolence (<1% in 1 mg group vs. 3% in 2 mg group), ataxia (<1% in 1 mg group vs. 3% in 2 mg group and flatulence (0% in 1 mg group vs. 3% in 2 mg group).

Add-on therapy

Adverse reactions reported with a higher incidence in placebo-controlled studies in patients receiving 1 mg/day rasagiline as add-on therapy to levodopa are presented in the table below with the adverse reaction incidence (% of patients) in rasagiline vs. placebo, respectively.

AEs in patients taking levodopa

		Rasagiline 1 mg (n = 380) %	Placebo (n = 388) %
Body as whole	Accidental injury	8.2	5.20
	Abdominal pain	3.9	1.3
	Neck pain	1.6	0.5
Cardiovascular System	Postural hypotension	4.7	1.3
	Angina pectoris	0.5	-
	Cerebrovascular accident	0.5	0.3
Digestive System	Constipation	4.2	2.1
	Dry mouth	3.4	1.8
	Vomiting	3.4	1.0
	Anorexia	2.1	0.5
Musculoskeletal system	Arthralgia	3.2	1.3
	Tenosynovitis	1.3	-
Metabolic and nutritional disorders	Weight loss	4.2	1.5
Nervous system	Dyskinesia	10.3	6.4
	Dystonia	2.4	0.8
	Abnormal dreams	2.1	0.8
	Ataxia	1.3	0.3
Skin and appendages	Rash	2.6	1.5
	Skin melanoma	0.5	0.3

- Serious adverse event/deaths/other significant events

No deaths were reported during the placebo-controlled monotherapy studies. Four out of a total 544 rasagiline-treated (0.5 mg and 1 mg) patients (0.7%), 4/388 (1%) placebo-treated patients and 3/227 (1.3%) entacapone-treated patients died during the placebo-controlled, LARGO and PRESTO adjunct therapy studies.

In the placebo-controlled studies, SAEs for the monotherapy cohort were slightly increased in the rasagiline groups vs. placebo (6 patients/ 4% for 1 mg, 9 patients/ 6% for 2 mg, and 4 patients/ 3% for placebo), with most individual AEs occurring in a single patient. In the 1 mg group, 4 of the 6 had

cardiovascular SAEs, including 2 with angina pectoris, one with intra-operative myocardial infarction and one with atrial fibrillation. One patient on 2 mg had angina pectoris and was discontinued due to need for medication that was not allowed.

For the adjunct therapy cohort, incidence of SAEs was the same for rasagiline and placebo treated patients (8%). Although the incidence of SAEs is twice that of the 1 mg group of monotherapy cohort, these patients were generally more complex medically with a longer history of PD and chronic levodopa treatment and the similarity of the rasagiline treated patients to placebo bears this out.

The SAEs reported are mostly cardio or cerebrovascular in origin they are in line with the age and the prevalence of the diseases in the population studied. There is no evidence that rasagiline contributed to their occurrence.

Melanoma

During the entire clinical program of rasagiline 20 cases of melanoma, most of them in situ, were reported in 19 patients (both prior to and after treatment initiation). Fifteen of the patients were treated with rasagiline, one patient was on placebo and additional 3 were diagnosed before treatment initiation. Eleven (11) of the 15 patients were diagnosed in studies without a concurrent placebo group. Six cases were diagnosed before the applicant has implemented active dermatological examinations in all ongoing clinical trials worldwide. Indeed, during the conduction of rasagiline clinical trials in North America two cases of invasive melanoma and four cases of melanoma in-situ were diagnosed. Despite the uncertain significance of this, the FDA thought it appropriate to institute additional surveillance. The applicant complied with this request by implementing periodical skin exams in all ongoing clinical trials. Therefore, it was not unexpected that following this step the number of diagnosed cases would rise.

The finding of 6 melanoma cases that were mostly appearing in rasagiline treated patients within its clinical development programme stirred a lot of scientific discussion to clarify the issue: there were more cases in rasagiline treated patients than in patients receiving placebo, even if there were also more patients in the programme exposed to rasagiline than to placebo. The prospective screening imposed by FDA determined, as expected, the finding of an extra number of cases. The epidemiological and clinical effort put in the clarification of this matter is an add-value of the rasagiline clinical programme.

In fact melanoma and PD have always been linked by the theoretical fear, based on the pharmacological mechanism, that LD would increase melanoma appearance or development, but no robust data were available. Nevertheless, epidemiological studies, some of them conducted independently from the applicant (e.g. in Denmark) show that PD patients have overall a lower risk of cancer but a higher risk of melanoma. The prospective studies also show that cancer registers underestimate the rates of melanoma in situ in the general population. The data available suggest that PD patients are at a higher risk for melanoma. The screening imposed by FDA show that melanomas in the rasagiline clinical programme occur at baseline before any exposure and do not increase with the cumulative exposure. Furthermore there is an imbalance of the number of skin lesions at baseline penalizing rasagiline groups, which reflects later on in the number of cases found in rasagiline or placebo, numerically superior in the rasagiline. The larger number of cases in US as compared with Europe is in line with the epidemiologic surveys.

In conclusion the epidemiologic data descriptive and analytic plus the screening data strongly suggested that melanoma is a problem related with the disease and not with rasagiline. Still this should be a topic for special monitoring within the Periodic Safety Update Reports during postmarketing and it should be advised in the SPC that any suspicious skin lesion should be evaluated by a specialist.

Weight loss

In the adjunct therapy studies, increased incidence of measured weight loss was noted in the rasagiline treatment groups as compared to placebo. Dose relationship was not seen in the PRESTO study. In the monotherapy TEMPO study, the incidence of measured weight loss >5% in both rasagiline treatment groups was very low and similar to placebo. Weight loss is likely to be an expected dopaminergic effect. Weight loss was reported as an AE with a low incidence (4.4%) for all patients ever treated with rasagiline. The problem of weight loss is intrinsic to Parkinson disease and is not very well understood. The safety database of rasagiline shows that weight loss is uncommon in early patients

(e.g. those in rasagiline monotherapy study). In the adjunct studies, weight loss was relatively frequent but it cannot be attributed to rasagiline since it happened similarly in the entacapone group. It is likely that weight loss has a complex origin dependent of the interaction of the disease process with dopaminergic therapies.

"Flu-like" syndrome

Analysis of the incidence and frequency of selected symptoms: flu syndrome, fever, malaise, arthritis and neck pain reported in the placebo-controlled phase of the TEMPO study shows that the adverse event (AE) "flu syndrome" was significantly higher in the rasagiline 1 mg group than in placebo, but without dose response relationship. The vast majority of these selected symptoms were reported as independent events and not in association with each other or with the COSTART term "flu syndrome". Most cases of "flu syndrome" in rasagiline treated patients were assessed by the investigator as mild, and none was severe.

With regard to possible immune system effects of rasagiline, the AE "infection" was reported equally in all groups: 14.9% for rasagiline 1 mg and 15.9% for both rasagiline 2 mg and placebo (TEMPO study). Severe AEs related to infection were reported similarly in each group: "infection" and "gastroenteritis", each in a rasagiline 2 mg and a placebo patient, and "abscess" in a rasagiline 1 mg patient. In order to further evaluate the relationship of rasagiline to "flu-like syndrome", a similar analysis was performed for the two pivotal studies in levodopa treated fluctuating PD patients, LARGO and PRESTO. A significant increase in "flu-like" syndrome in rasagiline treated patients was not demonstrated. Overall, the analysis does not suggest an effect of rasagiline on the immune system.

Neck pain was the only adverse reaction for which there was a dose dependent relationship.

Other important adverse events that were reported in clinical studies with rasagiline (other dose or in studies without placebo control) occurred in two patients each were rhabdomyolysis (both cases were following fall and prolonged immobilization) and inappropriate antidiuretic hormone (ADH) secretion. The complex nature of these cases makes it impossible to determine what role, if any, rasagiline played in their pathogenesis.

- Laboratory findings

Vital sign and ECG were studied.

Orthostatic hypotension is aggravated by rasagiline in add on therapy. The phenomenon was considered mild.

The scattered findings regarding QTc prolongation do not seem to indicate any worrying signal.

- Safety in special populations

Other than studies in renal impairment and hepatic impairment no relevant data arise. Hepatic and/or renal impairment might increase the rates of dopaminergic effects although the studies so far conducted did not detect such an effect.

- Safety related to drug-drug interactions and other interactions

Monoamine oxidase inhibitors (MAOI) are listed among agents that can induce the serotonin syndrome when co-administered with serotonin enhancing agents, including antidepressants such as the tricyclics, selective serotonin reuptake inhibitors (SSRIs), serotonin and noradrenaline reuptake inhibitors (SNRIs), trazodone, lithium and tryptophan. MAOIs combined with the opioids dextromethorphan and pethidine have also produced the serotonin syndrome. MAOI are assumed to contribute to this condition by preventing the metabolism of serotonin which is a preferential substrate of MAO-A, but can still be metabolized by MAO-B. Rasagiline is a potent, irreversible MAO-B selective inhibitor. Because of rasagiline's selectivity for MAO-B as compared to MAO-A, at the recommended clinical dose (1 mg/day), the risk of serotonin syndrome is limited. The concern regarding the possibility of occurrence of serotoninergic syndrome conducted to the avoidance of SSRI in the early phase of the development. In pivotal trials they were used but it happened in a limited way. Serotoninergic syndrome is a rare but serious condition and it is impossible to exclude a small risk for its occurrence thus warning has been included in SPC.

- Discontinuation due to adverse events

Overall, a small number of patients in the placebo-controlled studies discontinued prematurely due to AEs, suggesting that rasagiline is well-tolerated. Five patients on rasagiline 1 mg, two patients on 2 mg and one patient on placebo discontinued in the monotherapy cohort. All the AEs associated with termination were reported in only one patient, except for hallucinations which occurred in three rasagiline-treated patients (two on 1 mg, one on 2 mg). For the adjunct therapy cohort, approximately equal numbers of patients in the rasagiline (N=16, 4.2%) and placebo groups (N=19, 4.9%) terminated the study prematurely. Most AEs were recorded for only one patient. The body system with the highest incidence of AEs associated with premature discontinuation was the nervous system.

- Post marketing experience

Not applicable

- Discussion on clinical safety

The number of patients exposed to rasagiline in this clinical development plan is very close to 1500. The data generated show that rasagiline can be considered a well-tolerated drug. The most frequent adverse events in the lower doses (up to 1 mg/d) are unspecific but above this or when rasagiline 1 mg is added to levodopa they become clearly dopaminergic. Orthostatic hypotension might be problem in few patients, when rasagiline is used as add-on therapy.

The putative tyramine interaction was studied and described under pharmacodynamic studies.

During the clinical development programme the occurrence of cases of melanoma prompted the consideration of a possible association with rasagiline. The data collected suggests that Parkinson disease, rather than any drug in particular, is associated to a higher risk of melanoma.

5. Overall conclusions, benefit/risk assessment and recommendation

Quality

The quality of the product is considered to be acceptable when used in accordance with the conditions defined in the SPC. Physicochemical and biological aspects relevant to the uniform clinical performance of the product have been investigated and are controlled in a satisfactory way. There are no unresolved quality issues, which have a negative impact on the Benefit Risk balance of the product.

Non-clinical pharmacology and toxicology

Rasagiline was shown to be a potent, irreversible MAO-B selective inhibitor, which may cause an increase in extracellular levels of dopamine in the striatum.

Preclinical data reveal no special hazard for humans based on conventional studies of safety pharmacology, repeated-dose toxicity and reproduction toxicity.

Considering the equivocal genotoxic potential and the lack of plausible explanation for lung tumor development in mice, a further test addressing the involvement of a genotoxic mechanism in the lung tumorigenesis observed in mice has been requested. To further investigate the possibility of a mouse unique metabolite, which may be responsible for the results observed in the mutagenicity and carcinogenicity studies, the applicant initiated a study designed to compare rasagiline metabolites in mice and rats biofluids to those in humans. Although a mechanistic explanation is still lacking, the fact that tumours were only observed in mice reduces the concern regarding human safety.

Efficacy

The clinical development of rasagiline is based on 3 pivotal trials; one in monotherapy in early PD patients and 2 as add-on therapy to levodopa in patients with motor fluctuation. The 3 trials together demonstrate that rasagiline has a consistent anti-parkinson effect.

Treatment with rasagiline of early PD patients does produce a consistent beneficial effect measurable by the UPDRS total score and its sub-scores. The effect is modest (2.7 points on UPDRS motor score and 4.2 points on total UPDRS score) but is maintained in the long-term and is supported by other assessments like the responder analysis and Quality of Life which show a significant difference from placebo.

The 2 pivotal trials where rasagiline was tested as add-on to levodopa demonstrates that rasagiline reduces OFF time (1 h in more than 50% of the patients) and increases ON time without dyskinesias more that it increases ON with dyskinesias. The responder analysis for an improvement of at least 1.5h showed that about 40% of the patients reached this mark both for rasagiline and entacapone when the cut-off is put at the 2 hours, 30% of the actively treated patients reached it. The therapeutic indication should be restricted to patients with end of dose fluctuations as there were no data in other populations.

Safety

At the recommended dose (1 mg/d), rasagiline was well tolerated; the most frequent adverse events are unspecific in monotherapy. When rasagiline 1 mg is added to levodopa the most frequent adverse events are dopaminergic in nature (e.g. dyskinesia, postural hypotension and vomiting). Orthostatic hypotension in add-on therapy might also be a concern in few patients. Rasagiline should not be used in cases of moderate and severe hepatic impairment.

Due to known interactions between non selective MAO inhibitors and other medicinal products, specific recommendations for concomitant use of rasagiline with some other medicinal product have been included in the summary of product characteristics.

The most serious concern in the clinical development of rasagiline was the relation with an increased incidence of melanoma. The epidemiological studies conducted strongly suggest that the increased melanoma incidence is real but related to the disease rather than any product in particular. Recent epidemiological data also suggest that PD patients are at higher risk for skin cancers and not only melanoma.

6. Overall conclusions and benefit/risk assessment

Benefit/risk assessment

The benefit/risk of rasagiline is favourable:

- as monotherapy in early Parkinson's disease patients, where rasagiline has showed a modest effect with once daily administration without need for titration and is well tolerated,
- as adjunct therapy (with levodopa) in patients with end of dose fluctuations, where rasagiline reduces OFF time with an acceptable dopaminergic safety profile.

Recommendation

Based on the CHMP review of data on quality, safety and efficacy, the CHMP considered by consensus that the benefit/risk balance of AZILECT in the treatment of idiopathic Parkinson's disease (PD) as monotherapy (without levodopa) or as adjunct therapy (with levodopa) in patients with end of dose fluctuations was favourable and therefore recommended the granting of the marketing authorisation.