11 December 2025
EMA/CHMP/130428/2026
Committee for Medicinal Products for Human Use (CHMP)

Withdrawal Assessment report

Blarcamesine Anavex

International non-proprietary name: Blarcamesine

Procedure No. EMEA/H/C/006475/0000

Note

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

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List of abbreviations

Abbreviation	Definition
Aβ	Amyloid beta (also Abeta)
Aβ40	Amyloid beta isoform 40 (also Abeta40)
Aβ42	Amyloid beta isoform 42 (also Abeta42)
AD	Alzheimer's Disease
ADAS-Cog13	Alzheimer's Disease Assessment Scale-Cognitive Subscale
ADCS-ADL	Alzheimer's Disease Cooperative Study – Activities of Daily Living Scale
ADL	activities of daily living
ADRDA	Alzheimer's Disease and Related Disorders Association
AE	Adverse event
ANAVEX2-73	blarcamesine
API	Active Pharmaceutical Ingredient
APP	amyloid precursor protein
ARIA	Amyloid-Related Imaging Abnormalities
ASM	Active Substance Manufacturer
AST	aspartate aminotransferase
BADL	Basic activities of daily living
BCS	Biopharmaceutics Classification System
BL	baseline
BMI	body mass index
BTD	best tolerated dose
CCB	Cogstate Brief Battery
CDR-SB	Clinical Dementia Rating Scale Sum of Boxes
CFB	Change from baseline
cfu	colony forming unit
CI	Confidence interval
CGI-I	Clinical Global Impression – Improvement
CGI-S	Clinical Global Impression – Severity
CoA	Certificate of Analysis
Conc	concentration group
COVID-19	*Corona virus* disease-2019
CQA	Critical Quality Attribute
CRS	Chemical Reference Substance (official standard)
CSF	cerebrospinal fluid
CSR	clinical study report
C-SSRS	Columbia-Suicide Severity Rating Scale
CT	Computed tomography
CTCAE	Common Terminology Criteria for Adverse Events
CYP	cytochrome P450
DB	double blind
DDI	drug-drug interaction
Diff	Difference
DMF	Drug Master File = Active Substance Master File
DP	Decentralised (Application) Procedure
DSC	Differential Scanning Calorimetry
DSM-IV	Diagnostic and Statistical Manual of Mental Disorders, Fourth Edition

EEG	Electroencephalogram
EEP	Efficacy Evaluable Population
EOS	End-of-study
ER	Exposure-response
ERP	Event-Related Population
FAS	Full Analysis Set
FCSRT	Free and Cued Selective Reminding Test
FDG	Fluorodeoxyglucose
FDG-PET	Fluorodeoxyglucose-positron emission tomography
GLM	General linear model
GPCR	G protein-coupled transporter
HAM-D	Hamilton Psychiatric Rating Scale for Depression
HDPE	High Density Polyethylene
HPLC	High Pressure Liquid Chromatography
IADL	Instrumental activities of daily living
ID	identification number
IMM	Independent Medical Monitor
IPC	In-process control test
IR	Infrared
ISLT	International Shopping List Task
ITT	Intent-to-Treat
LLN	lower limit of normal
LOD	Limit of Detection
LOQ	(1) Limit of Quantification, (2) List of Questions
LS	Least squares
M	medium
MA	Marketing Authorisation
mAb	monoclonal antibody
MAD	multiple ascending dose
MAH	Marketing Authorisation holder
MAR	Missing at random
Max	maximum
MCI	mild cognitive impairment
MCV	mean corpuscular volume
MedDRA	Medical Dictionary for Regulatory Activities
Min	minimum
MMRM	Mixed-effects model of repeated measurements
MMSE	Mini-Mental State Examination
MNAR	Missing not at random
MoA	Mechanism of Action
MRI	magnetic resonance imaging
MS	Mass Spectrometry
MTD	maximal tolerated dose
N/A	not available
NA	Not applicable
ND	Not detected
NfL	neurofilament light chain
NIA-AA	National Institute on Aging and Alzheimer's Association
NINCDS	National Institute of Neurological and Communicative Disorders and

	Stroke
NMDA	N-methyl-D-aspartate
NMR	Nuclear Magnetic Resonance
NMT	Not more than
NPI-Q	Neuropsychiatric Inventory-Questionnaire
OL	open-label
OLE	Open-label extension
OOS	Out of Specifications
PCB	placebo
PDE	Permitted Daily Exposure
PE	Polyethylene
PET	positron emission tomography
PI	Principal Investigator
PP	Polypropylene
PSD	Particle size distribution
PSEN1	presenilin 1
PSEN2	presenilin 2
PT	preferred term
p-Tau	phosphorylated Tau
PVC	Poly vinyl chloride
QC	Quality Control
QD	once daily
QOS	Quality Overall Summary
QP	Qualified Person
QTcB	QT interval corrected using Bazett's formula
QTcF	QT interval corrected using Fridericia's formula
RH	Relative Humidity
RP	Restricted Part (or Closed Part) of a DMF
RRT	Relative retention time
RSD	Relative standard deviation
S1R/SIGMAR1	Sigma-1 receptor
S2R	Sigma-2 receptor
SAE	Serious adverse event
SAP	Statistical Analysis Plan
SD	Standard deviation
SE	Standard error
SMCC	Silicified Microcrystalline Cellulose
SOC	System Organ Class
TEAE	Treatment-emergent adverse event
TGA	Thermo-Gravimetric Analysis
TOEP	Standard Toeplitz
TOEPH	Heterogeneous Toeplitz
ULN	upper limit of normal
UN	Unstructured
UPLC	Ultra Pressure Liquid Chromatography
UV	Ultraviolet
vs.	versus
WBC	white blood cell
wk	week
WT	Wild type

XRPD	X-ray powder diffraction (= PXRD)

1. Background information on the procedure

1.1. Submission of the dossier

The applicant Anavex Germany GmbH submitted on 23 November 2024 an application for marketing authorisation to the European Medicines Agency (EMA) for Blarcamesine Anavex, through the centralised procedure falling within the Article 3(1) and point 1 of Annex of Regulation (EC) No 726/2004.

The applicant applied for the following indication: add-on therapy for the treatment of early Alzheimer's disease in adults with mild cognitive impairment (MCI) due to Alzheimer's disease or early-stage mild dementia due to Alzheimer's disease, in patients with SIGMAR1 wild-type genotype.

1.2. Legal basis, dossier content

The legal basis for this application refers to:

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

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

The applicant requested during the procedure consideration of its application for a Conditional Marketing Authorisation in accordance with Article 14(a) of Regulation (EC) No 726/2004.

1.3. Information on Paediatric requirements

Pursuant to Article 7 of Regulation (EC) No 1901/2006, the application included an EMA Decision CW/1/2015 on the granting of a class waiver.

1.4. Information relating to orphan market exclusivity

1.4.1. Similarity

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

1.5. Applicant's request(s) for consideration

1.5.1. Conditional marketing authorisation

The applicant requested consideration of its application for a Conditional marketing authorisation in accordance with Article 14(a) of Regulation (EC) 726/2004.

1.5.2. New active Substance status

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

1.6. Scientific advice

The applicant did not seek Scientific advice from the CHMP.

1.7. Steps taken for the assessment of the product

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

Rapporteur: Janet Koenig Co-Rapporteur: Ewa Balkowiec Iskra

The application was received by the EMA on	23 November 2024
The procedure started on	27 December 2024
The CHMP Rapporteur's first Assessment Report was circulated to all CHMP and PRAC members on	17 March 2025
The PRAC Rapporteur's first Assessment Report was circulated to all PRAC and CHMP members on	28 March 2025
The CHMP agreed on the consolidated List of Questions to be sent to the applicant during the meeting on	25 April 2025
The applicant submitted the responses to the CHMP consolidated List of Questions on	19 July 2025
The following GCP inspection was requested by the CHMP and their outcome taken into consideration as part of the Quality/Safety/Efficacy assessment of the product:	
– A GCP inspection at the Sponsor site in US and at two clinical sites, located in Australia and Germany between 28 April 2025 and 31 May 2025. The outcome of the inspection carried out was issued on 15 August 2025	15 August 2025
The CHMP Rapporteurs circulated the CHMP and PRAC Rapporteurs Joint Assessment Report on the responses to the List of Questions to all CHMP and PRAC members on	25 August 2025
The PRAC agreed on the PRAC Assessment Overview and Advice to CHMP during the meeting on	04 September 2025
The CHMP agreed on a list of outstanding issues in writing and/or in an oral explanation to be sent to the applicant on	18 September 2025
The applicant submitted the responses to the CHMP List of Outstanding Issues on	14 October 2025

The CHMP Rapporteurs circulated the CHMP and PRAC Rapporteurs Joint Assessment Report on the responses to the List of Outstanding Issues to all CHMP and PRAC members on	11 September 2025
The outstanding issues were addressed by the applicant during an oral explanation before the CHMP during the meeting on	11 November 2025
The CHMP, in the light of the overall data submitted and the scientific discussion within the Committee, issued a negative opinion for granting a marketing authorisation to Blarcamesine Anavex on	11 December 2025
Furthermore, the CHMP adopted a report on New Active Substance (NAS) status of the active substance contained in the medicinal product	11 December 2025

2. Scientific discussion

2.1. Problem statement

2.1.1. Disease or condition

Alzheimer's Disease (AD) is a progressive neurodegenerative disorder characterized cognitive decline, memory impairment, and changes in behaviour and personality. It is the most common cause of dementia among older adults, accounting for approximately 60-80% of all dementia cases (Alzheimer's Association).

Women continue to be disproportionately affected by dementia with 6,650,228 women and 3,130,449 men living with dementia in Europe. The disease primarily affects individuals over the age of 65, although early-onset Alzheimer's can occur in individuals as young as their 30s or 40s, albeit rarely.

Patients go through a stage of Mild Cognitive Impairment and subsequently deteriorate into progressively severe dementia. AD is the most common cause of dementia among older adults and is ultimately fatal.

2.1.2. Epidemiology and risk factors

Currently more than 55 million people are suffering from dementia worldwide, over 60% of whom live in low- and middle-income countries. The number of new cases is 10 million each year (WHO 2023). The number of people living with dementia is estimated to be Approximately 7.8 million people live with dementia in the European Union (EU27), while 9.8 million people are estimated in European countries represented by Alzheimer Europe members (Alzheimer Europe 2023).

Risk factors for AD basically include older age, genetics (especially the e4 form of the APOE gene), and a family history of AD (Alzheimer’s Association 2022; Alzheimer Europe 2019). Less than 5% of all AD cases exhibit an autosomal dominant inheritance pattern (Wu et al. 2012). The remaining 95% are the non-dominantly inherited type known as sporadic AD, in which the APOE ε4 acts as the strongest genetic risk factor (Alzheimer’s Association 2022). Factors that increase the risk of cardiovascular disease are also associated with a higher risk of sporadic AD (Roberts and Knopman 2013; Alzheimer’s Association 2022), i.e. active smoking and diabetes (Anstey et al. 2007; Durazzo et al. 2014; Lee et al. 2018), as well as high serum cholesterol, hypertension, and obesity, particularly in midlife (Anstey et

al. 2011; Meng et al. 2014; Anstey et al. 2017; Lennon et al. 2019). Other factors such as physical activity, healthy diet, more years of education, and being socially and mentally active are associated with a reduced AD risk (Chen et al. 2016; Rege et al. 2017; Alzheimer's Association 2021).

2.1.3. Aetiology and pathogenesis

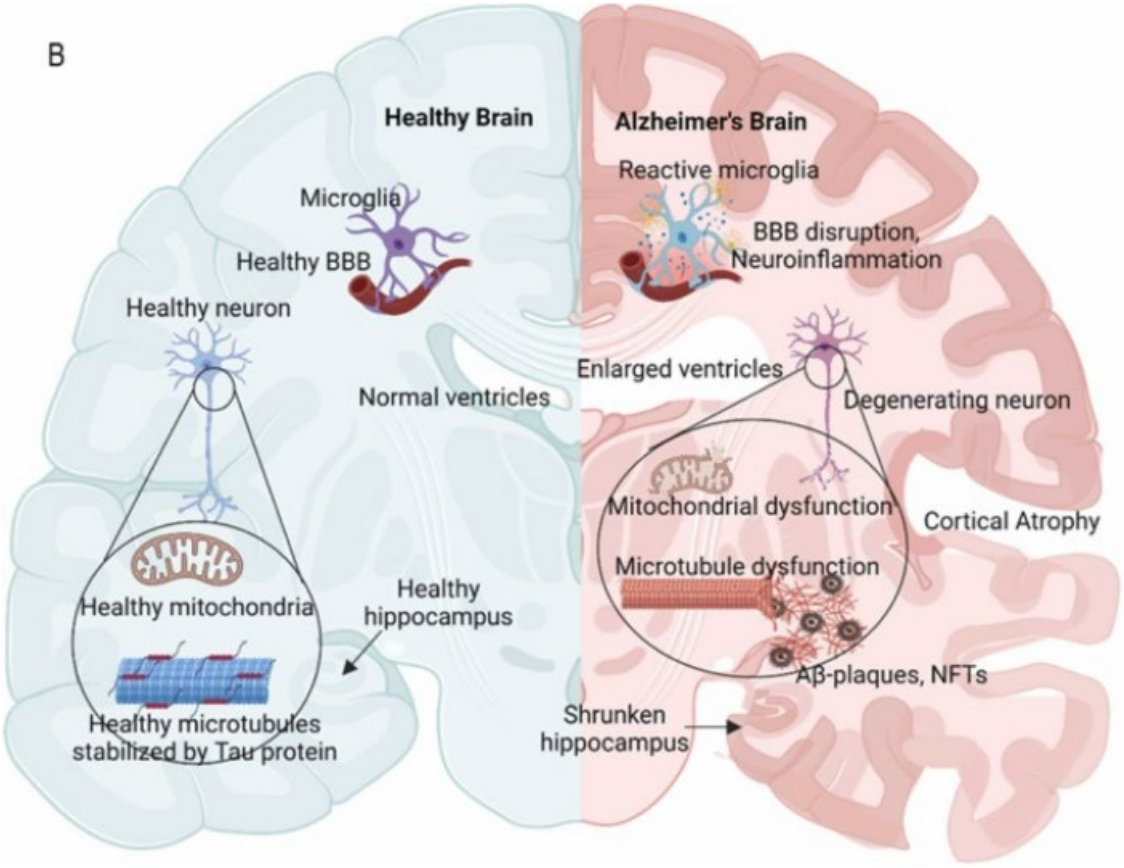
AD is associated with neuropathological changes in the brain such as deposits of beta-amyloid and tau which are likely important components of the pathophysiological process. However, the relationship remains far from fully understood. Moreover, these changes are seen even in persons who are without symptoms related to AD dementia and may occur many years prior to disease-onset. Pathologically, Alzheimer's disease is characterized by the accumulation of two types of abnormal protein aggregates in the brain: beta-amyloid plaques and tau tangles. Beta-amyloid plaques are extracellular deposits formed by the aggregation of beta-amyloid protein fragments derived from the amyloid precursor protein (APP). Amyloid disease can be broadly classified as familial AD (FAD) and sporadic AD (SAD). Tau tangles, on the other hand, are intracellular accumulations of hyperphosphorylated tau protein, which disrupts the normal functioning of neurons and ultimately leads to cell death (Kumar 2015).

The exact aetiology of Alzheimer's disease remains incompletely understood, but it is believed to involve a complex interplay of genetic, environmental, and lifestyle factors. While age is the greatest risk factor for developing Alzheimer's disease, individuals with a family history of the condition are also at increased risk. Additionally, certain genetic mutations, such as those in the genes encoding APP, presenilin 1 (PSEN1), and presenilin 2 (PSEN2), have been linked to early onset familial forms of the disease (Hampel 2021).

According to the literature, the preclinical phase of Alzheimer's disease is the cellular phase. Alterations in neurons, microglia, and astroglia drive the insidious progression of the disease before cognitive impairment is observed. Neuro-inflammation, alterations in the vessels, ageing, and dysfunction of the glymphatic system act upstream or in parallel to accumulating amyloid β in this cellular disease landscape. Tau tangles are positively correlated with the severity of AD and associated with cognitive impairment. Amyloid β induces, via an unknown pathway, the spread of tau pathology which is associated with the appearance of necroptosis markers in neurons displaying granulovacuolar degeneration (Zhang et al., 2023, Scheltens et al., Alzheimer's disease, Lancet 2021).

The current understanding of the mechanisms involved in pathogenesis has been summarized by Khan et al. (Recent Advancements in Pathogenesis, Diagnostics and Treatment of Alzheimer's Disease, Current Neuropharmacology 2020). Complex interplay of various pathological mechanisms in AD are shown in Figure 1 below.

Figure 1. Alzheimer's disease and its pathology (from Kamatham et al., 2024)



Causative factors and hallmarks of AD. Several pathological mechanisms like neuroinflammation, mitochondrial dysfunction, oxidative stress, blood brain barrier disruption, protein aggregation play role in AD progression. Hallmarks include the presence of pathological proteins such as Aβ plaques, p-tau, neurofibrillary tangles (NFTs) and anatomical changes like enlarged ventricles, cortical atrophy, shrunken hippocampus.

2.1.4. Clinical presentation, diagnosis and prognosis

Both the underlying pathophysiological process of AD and its clinical symptomatology are best conceptualised as a continuum: patients progress from normal cognition to Mild Cognitive Impairment (MCI) due to AD, followed by increasing severity of AD dementia (mild, moderate, and severe).

MCI due to AD is a pre-dementia phase of AD, characterised by the development of noticeable memory problems (amnestic) or impaired judgment or decision-making (non-amnestic), which does not affect independence of functional abilities, does not meet the criteria for dementia, and has AD as a suspected aetiology (Davis et al., 2018).

During the progression of AD there are 3 broad phases: pre-clinical AD, Mild Cognitive Impairment (MCI) due to AD, and AD dementia (Alzheimer's Association 2021; Alzheimer Europe 2019). Updated IWG criteria (Dubois 2024) meanwhile enable a diagnosis of AD at an early prodromal stage according to the concept of a clinical-biological entity. Depending on the degree of symptoms interfering with the ability to carry out daily tasks, the AD dementia phase is further broken down into mild, moderate, and severe stages (Alzheimer's Association 2021; Alzheimer Europe 2019). Patients with mild AD dementia may be able to function independently in many daily tasks but is likely to require assistance with some

activities to maximise independence and remain safe (Alzheimer's Association 2021; Alzheimer Europe 2019).

Clinical symptoms of AD include progressive memory decline, impaired executive function and difficulties executing routine daily activity; early symptoms of AD onset include changes in thinking or unconscious behaviour, memory impairment with respect to new information, and dysfunctional changes in language and speech (Guo et al. 2020). Decline in memory has been considered the predominant and earliest symptom of AD, followed by impairments of other cognitive domains such as language, executive function, praxis, and complex visual processing, though heterogeneity in the presentation and course of cognitive impairments has been identified (Dubois et al. 2010). Behavioural and psychological symptoms have also been recognised as a key feature of AD dementia (Robert et al. 2005; Bature et al. 2017). Approximately 20 to 30% of early AD patients show significant depressive symptoms and mood changes. Patients in advanced stages of AD suffer from severe memory loss, hallucinations, disorientation, and lack self-sufficiency, where individuals eventually die due to respiratory syndrome, infection or fasting (Guo et al. 2020).

Individuals with MCI or AD dementia are at increased risk of mortality. Across studies, the HR of all-cause mortality among individuals with MCI ranged from 1.5 to 2.0 when compared to that among individuals without cognitive impairment (Guehne et al. 2007; Wilson et al. 2009; Vassilaki et al. 2015). A European study of death data in 28 countries estimated that the overall age-standardised mortality rate of AD increased from 28.2 to 45.2 per 100,000 people during 1994 to 2013 (Niu et al. 2017b).

Clinical assessment and cognitive screening are often the initial steps in evaluating individuals suspected of having AD. These steps involve gathering a comprehensive medical history, conducting physical examinations, and assessing cognitive function through various standardised tests.

2.1.5. Management

At present, there is no cure for AD, and existing treatments focus primarily on managing symptoms and slowing disease progression. In the European Union, approved therapies for AD include cholinesterase inhibitors (e.g. donepezil, rivastigmine, galantamine) for patients with mild to moderately severe AD and the non-competitive N-methyl-D-aspartate receptor antagonist memantine for patients with moderate to severe AD. These medications provide modest symptomatic relief but do not address the underlying pathology of the disease (Birks J, 2006).

Cognitive dysfunctions in Alzheimer's disease are explained by the loss of cholinergic neurons Thus, acetylcholinesterase (AChE) inhibitors are used as interventions to prevent the hydrolysis of acetylcholine by the enzyme acetylcholinesterase, which leads to increased acetylcholine levels in the synaptic cleft and improved neurotransmission (Kamatham et al 2024).

Several earlier clinical studies with anti- amyloid beta (Aβ) monoclonal antibodies as disease-modifying treatments (bapineuzumab, solanezumab, crenezumab and gantenerumab) did not meet their clinical endpoints. Recently, the MAA for aducanumab was withdrawn since the CHMP reached a negative opinion because the pharmacodynamic effect (i.e., reduction of brain amyloid load) did not translate into a clinical effect. In November 2024, the CHMP adopted a positive opinion for the monoclonal Aβ antibody lecanemab in the treatment of mild cognitive impairment and mild dementia in adults with AD, who are non-carriers or heterozygotes of apolipoprotein E ε4 with confirmed amyloid pathology ("Leqembi"; EMEA/H/C/5966). The European Commission authorised Leqembi in April 2025. Moreover, another monoclonal antibody (donanemab, trade name: "Kisunla") received marketing authorisation in the UK for the same clinical indication. In July 2025 CHMP adopted a positive opinion, recommending the granting of a marketing authorisation for the medicinal product Kisunla, intended for the treatment

of early symptomatic Alzheimer's disease in adults who are apolipoprotein E ε4 (ApoE ε4) non-carriers or heterozygotes. In September 2025 European Commission issued the decision for the approval of Kisunla's marketing Authorisation.

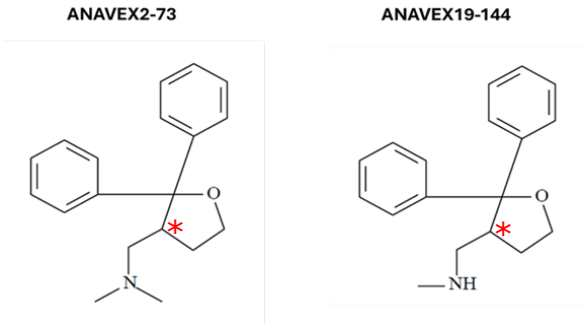
At the time of writing this report, it is premature to conclude on the impact of the recently approved amyloid-targeting antibodies on the management of AD patients in the EU.

2.2. About the product

Blarcamesine (also termed ANAVEX2-73 in the dossier), an aminotetrahydrofuran derivative, is a small molecule agonist of the Sigma 1 receptor (S1R) and, to a lower extent, a modulator of the muscarinic acetylcholine receptor (M1 to M5) that is proposed for treatment of early AD in adults.

Activation of S1R in the central nervous system (CNS) promotes neuroprotection by enhancing intracellular signalling pathways involved in the induction of cellular waste autophagy, mitochondrial function, and calcium homeostasis (Figure 3). The modulation of S1R also exerts anti-inflammatory effects and reduces oxidative stress in the endoplasmic reticulum (ER). These neuroprotective activities, which are shared by the structurally related demethylated active metabolite ANAVEX19-144 (Figure 2), are thought to indirectly counter two pathologies of AD, the burden of amyloid beta (Aβ) and the hyperphosphorylation of tau, which are implicated in the clinical deficits of AD.

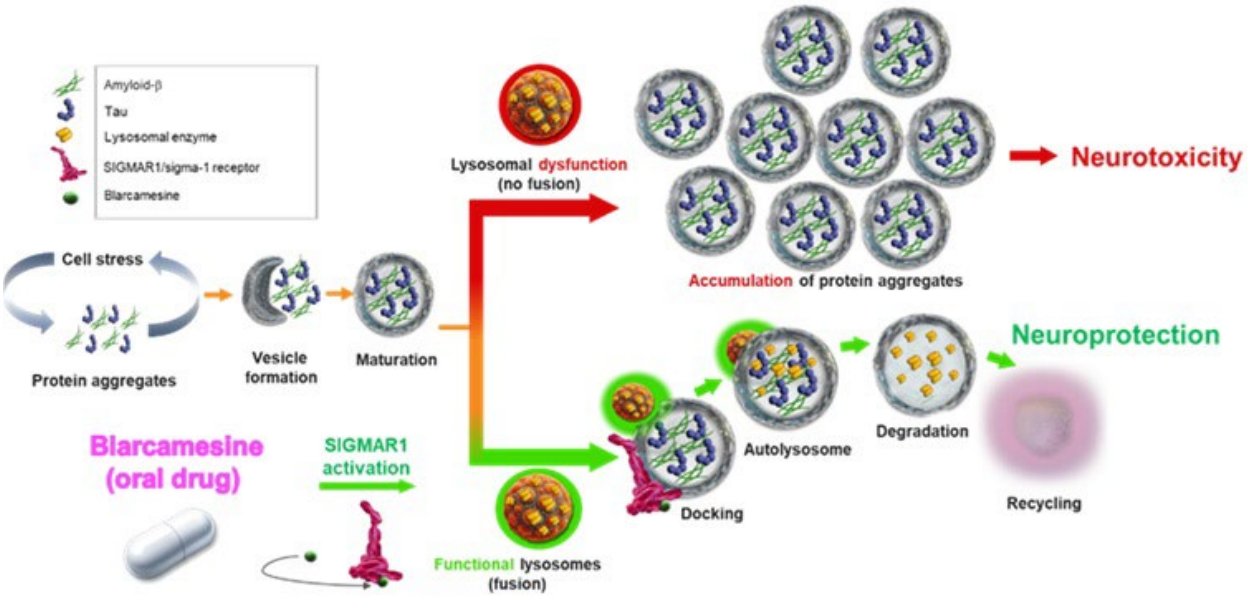
Figure 2. Chemical structures of blarcamesine (ANAVEX2-73) and its demethylated active metabolite (ANAVEX19-144)



The red asterisk denotes the single chiral centre contained in parent compound and metabolite; compiled based on the data in Modules 2.3 and 2.4

The active substance blarcamesine hydrochloride contains one chiral centre and is a racemate of equal amounts of R- and S-enantiomers with closely similar binding affinities for the tested cellular receptors.

Figure 3. Mechanistic activity of blarcamesine and its active metabolite ANAVEX19-144



Blarcamesine is not yet approved for use in any market.

The following indication was originally proposed: "*Blarcamesine is indicated for the treatment of Alzheimer's disease and dementia in adults*" and has subsequently been adapted to "*treatment of early Alzheimer's disease in adults with mild cognitive impairment (MCI) due to Alzheimer's disease or early-stage mild dementia due to Alzheimer's disease"* and is meanwhile been proposed "*as add-on therapy for the treatment of early Alzheimer's disease in adults with mild cognitive impairment (MCI) due to Alzheimer's disease or early-stage mild dementia due to Alzheimer's disease, in patients with SIGMAR1 wild-type genotype*".

Blarcamesine has been formulated as immediate-release hard gelatine capsule consisting of a binary blend of 11.3 mg blarcamesine hydrochloride (corresponding to 10 mg blarcamesine) and 73.7 mg silicified microcrystalline cellulose (PROSOLV® SMCC 50). The capsules should be swallowed whole after dinner.

The recommended oral starting dose for treatment of AD and dementia in adults is 10 mg once daily. Depending on the individual tolerability, this dose should be gradually titrated. Dose increments, if needed, should not exceed 10 mg at intervals of no less than 2 weeks (preferably 3 to 4 weeks), based on individual patient tolerability up to a maintenance and also maximum dose of 30 mg.

2.3. Type of application and aspects on development

The applicant requested consideration of its application for a Conditional Marketing Authorisation in accordance with Article 14(a) of Regulation (EC) 726/2004 based on the following claims according to the Regulation (EC) No 507/2006 on the conditional marketing authorisation for medicinal products for human use:

- That there is a positive benefit-risk balance;
- That it is likely that the applicant will be able to provide comprehensive data: the applicant refers to a confirmatory, large-scale clinical program;

- That unmet medical needs will be addressed, as the applicant claimed there is a need for oral medicines like blarcamesine;
- That the benefits to public health of the immediate availability outweigh the risks inherent in the fact that additional data are still required. Here the applicant again focused on the accessibility of what would an oral treatment provides patient-centred advantages, and then claimed that appropriate risk mitigation would be based on restricting the indication to a genetically-defined population, on an enhanced pharmacovigilance program, on prescriber education and real-world monitoring.

2.4. Quality aspects

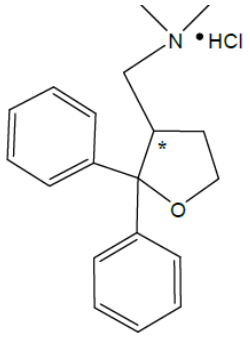
2.4.1. Introduction

The finished product is presented as hard capsules containing 10 mg of blarcamesine (as blarcamesine hydrochloride) as active substance.

2.4.2. Active Substance

General information

The chemical (IUPAC) name of blarcamesine hydrochloride is 1-(2,2-diphenyloxolan-3-yl)-N,N-dimethylmethanamine hydrochloride, which corresponds to the molecular formula $C_{19}H_{23}NO.HCl$. It has a relative molecular mass of 317.85 g/mol and the following structure:



* Chiral center

Figure 4. Active substance structure

The chemical structure of the active substance was elucidated by a combination of elemental analysis, mass spectroscopy, UV- and IR-spectroscopy, ^{1}H-NMR spectroscopy and ^{13}C-NMR spectroscopy supported by COSY, DEPT, HMBC and HSQC spectra and single crystal X-ray analysis. The solid state properties of the active substance were measured by XRPD, DSC, DVS and TGA.

The active substance is a non-hygroscopic white to off-white crystalline solid with a melting point of 227.6 °C and soluble in methanol and chloroform. The solubility in media of the physiological pH range is pH dependent: 132.7 mg/mL in pH 1.5, 148.4 mg/mL in pH 4.5 and 0.6 mg/mL in pH 6.9 (at 25°C).

It is manufactured in a racemic crystal (Form I), containing both enantiomers (R/S isomers) in the same crystal lattice in equal amounts.

Blarcamesine hydrochloride exists in eight polymorphic forms, denoted as forms I through VIII. Form I is thermodynamically preferred racemic crystal form, and it is also the target active substance crystal form.

Manufacture, characterisation and process controls

Detailed information on the manufacturing of the active substance has been provided in the restricted part of the ASMF and it was considered satisfactory.

The active substance is manufactured at two manufacturing sites. Satisfactory GMP documentation has been provided.

The manufacturing process of the active substance consists of five synthetic steps.

Specification

The active substance specification includes tests for identity (visual, IR, HPLC), water (Ph. Eur.), residue on ignition (Ph. Eur.), assay (HPLC), chemical purity (HPLC), tosylates (HPLC), dimthylamine hydrochloride (IC), residual solvents (GC-HS), sum of genotoxic impurities (calculation), particle size distribution (laser light scattering), and crystal modification (XRPD).

The specification of the active substance contains relevant parameters and justifications of specification have been provided. Tests and acceptance limits for standard compendial test methods are performed in accordance with the current requirements. For non-compendial specific tests, such as assay, impurities and residual solvents, ICH Q6A, ICH Q3A, ICH Q3C guidance documents were used as guide. The absence of control of microbial purity has been justified based on ICH Q6A guideline, Decision Tree #6.

Sufficiently detailed description of analytical procedures used for analysing active substance has been provided.

Impurities present at higher than the qualification threshold according to ICH Q3A were qualified by toxicological and clinical studies and appropriate specifications have been set.

All in-house methods have been adequately validated. The validation reports of the analytical test methods have been provided supported.

Reference and working standards of the active substance have been described and batch analysis data presented along with characterisation data. Impurity reference standards are used for quantitation.

Certificates of analysis of have been provided. The results are within the specifications and consistent from batch to batch.

Stability

Stability data from registration stored in the intended commercial package for up to 24 months (2 batches) and 12 months (3 batches) under long term conditions (25 ºC / 60% RH) and for up to 12 months under accelerated conditions (40 ºC / 75% RH) according to the ICH guidelines were provided.

Additional supporting stability data of ongoing studies is available.

The following parameters were tested during the stability studies performed on the five milled batches: appearance, assay, impurities, water content, crystal modification, and microbial limits.

No significant trends or changes in any test parameter investigated are observed.

Forced degradation testing was performed on one batch of the active substance. Degradation was achieved under the photolytic and oxidative conditions, no degradation was observed with heat, basic and acidic conditions.

The stability results indicate that the active substance manufactured by the proposed manufacturers is sufficiently stable. The stability results justify the proposed retest period of 24 months (2 years) without defined storage conditions in the proposed container.

2.4.3. Finished Medicinal Product

Description of the product and Pharmaceutical development

The proposed finished product is presented as a white to off-white hard gelatine capsule size 19.4 × 6.9 mm.

Particle size and polymorphic form are also described as two active substance properties that are critical for the manufacturability of the finished product and are controlled in the final active substance specification with suitable limits. Regarding particle size adequate optimization studies have been performed. Based on the provided results for these studies the proposed particle size distribution (PSD) specification for the active substance as included in the active substance specification is considered justified.

In relation to polymorphic form, there are eight polymorphic/pseudo-polymorphic forms discovered to date. Forms I, II and III are the only forms found to be relevant to the synthesis of the active substance. Form I is the target active substance form and has been consistently obtained to date during active substance manufacturing. Form II readily loses water upon heating above ambient temperature to collapse into Form III and has never been observed. The finished product manufacturing process was designed to avoid the conditions known to promote conversion of Form I to Form III.

During formulation development, a study was performed where a binary mixture of silicified microcrystalline cellulose and active substance with and without water was stored at ICH accelerated conditions for 30 days. The results of this study showed that the active substance with silicified microcrystalline cellulose mixture as the excipient is stable.

All excipients are well known pharmaceutical ingredients, and their quality is compliant with Ph. Eur. standards. There are no novel excipients used in the finished product formulation.

The pharmaceutical development has been designed to produce immediate release product with a simple formulation that could be filled into capsules. Initially, formulation development focused on a potential dose range. The 10 mg strength was chosen for commercialisation.

A quality target product profile (QTPP) was developed for the finished product to assure a safe, efficacious, and convenient dosage form. This profile along with the finished product critical quality attributes are provided

The development of the dissolution method was performed and was based on the solubility properties of the active substance in different media of the physiological range. The media where sink conditions were met were chosen for development of the dissolution method. Comparative dissolution data and profiles for one clinical batch of the finished product produced are provided and show more than 85% (mean) release after 15minutes in all relevant media.

The discriminatory nature of the dissolution method has been demonstrated by using batches with and

without compaction during encapsulation and by evaluating two different introduction modes. The presented evaluation of the discriminatory property of the dissolution method is supported with corresponding dissolution data.

The manufacturing process development consists of simple steps (blending, encapsulation, blistering). The process was initially developed using manual capsule filling in one manufacturing site, the phase 2 clinical trial and the pivotal clinical trial batches were then produced with batch sizes up to in another manufacturing site. The registration batches were produced at the commercial manufacturing site. As the manufacturing process is quite simple, additional optimization of the process after transfer to the commercial site was not performed. The different manufacturing sites used during product development are described in sufficient detail. A comparison of the manufacturing process parameters for the 10 mg capsule at the three manufacturing sites has been provided. The presented description of the manufacturing process is considered sufficient as the manufacturing process is very simple.

The primary packaging is PVC/PE/PCTFE blister packs with laminated aluminium foil backing. The material complies with Ph. Eur. and EC requirements. The choice of the container closure system has been validated by stability data and is adequate for the intended use of the product.

Manufacture of the product and process controls

The finished product is manufactured by one manufacturing site. Satisfactory evidence of GMP compliance has been provided for all sites involved in the manufacturing, testing and batch release of the finished product.
The manufacturing process consists of 5 main steps: blending and milling of a single excipient silicified microcrystalline cellulose with the active substance, encapsulation in hard gelatine capsules and blister packaging of the capsules. The process is considered to be a standard manufacturing process.

Process validation of the manufacturing process has not been performed as the manufacturing process is simple and does not belong to non-standard processes. A validation scheme to be followed for the validation of the commercial scale batches is included where only a very general description of the validation is provided.

To be in line with EMA Guideline on process validation for finished products (EMA/CHMP/CVMP/QWP/BWP/70278/2012-Rev1, Corr.1), it has been requested as Other Concern (OC) that either a detailed process validation plan or process validation data of the validation batches should be provided. This applicant's response to this OC is still awaited.

Product specification

The finished product release and shelf-life specifications include appropriate tests for this kind of dosage form: appearance (visual), identity (HPLC-UV), assay (HPLC-UV), content uniformity (Ph. Eur.), related substances (HPLC-UV), dissolution (Ph. Eur.), water content (KF), and microbiological test (Ph. Eur.)

The proposed specification is justified with reference to current EMA guidelines (ICH Q6A, ICH Q3B, ICH Q3D) and currently available batch analytical and stability data. Moreover, justification is provided for not including test on polymorphism into the finished product specification. The presented justification is supported with comparative of initial time point and from the end of stability of the five clinical batches included in the stability programme. Based on the presented data absence of test on polymorphism can be accepted.

For characterisation of impurities reference is made to the active substance section. This is considered

acceptable as both known impurities in the finished product are also described in the active substance. There are no impurities or degradation products known to form during the finished product manufacture. Residual solvent testing is not performed on the finished product as there are no residual solvents in the manufacturing process.

Updated sections 3.2.P.5.2 and 3.2.P.5.3. have been requested by the CHMP.

The potential presence of elemental impurities in the finished product has been assessed following a risk-based approach in line with the ICH Q3D Guideline for Elemental Impurities. Potential impurities that may be introduced from the drug product components, manufacturing process, purified water used to clean the equipment, and packaging components were considered in the risk assessment. According to the presented calculation the 30% control threshold as per ICH 3D is not exceeded. Hence, additional controls for elemental impurities in the drug product are not required.

The nitrosamine risk assessment report provided includes relevant information regarding potential risks connected to the active substance, excipients, capsules shells and packaging material. The risk evaluation by the applicant concluded that there is low probability of risk for generation of nitrosamines, and therefore confirmatory testing was not performed. However, the active substance itself is a tertiary amine, and an impurity of the active substance (mono-methyl derivative ANA001-31) is a secondary amine. Further, dimethylamine is used in the penultimate step in the synthesis of the active substance. Dimethylamine is also included in the active substance specification with a limit of NMT 0.15%. Further, nitrocellulose is present in the primary packaging material. Hence, risk factors for formation of nitrosamines are present and consequently formation of nitrosamines cannot be ruled out unambiguously. Consequently, the confirmatory testing is considered necessary regarding nitrosamines originating from the impurity ANA001-31 (mono-methyl derivative of the active substance) and dimethylamine. Therefore, the CHMP requested to provide the confirmatory testing as Major Objection (MO). The Applicant stated they have received the confirmatory testing results for nitrosamine impurities NDSRI (originating from the impurity ANA001-31) and NDMA (dimethylamine) in both the active substance and finished product, and that no risk of nitrosamines was found. However, as no actual results or appropriate documentation to substantiate the claims by the Applicant have been provided, this was not enough to resolve the MO.

The analytical methods used have been adequately described and appropriately validated in accordance with the ICH guidelines. It is stated that same reference standards are used as for analysis of the active substance and summary of the analytical data of these standards are presented.

Batch analysis results are provided for batches confirming the consistency of the manufacturing process and its ability to manufacture to the intended product specification.

Stability of the product

Stability data from finished product stored for up to 12 months under long term conditions (25 ºC / 60% RH) and for up to 12 months under accelerated conditions (40 ºC / 75% RH) according to the ICH guidelines were provided. The batches are identical to those proposed for marketing and were packed in the primary packaging proposed for marketing.

Supporting stability at ICH long term and accelerated conditions was provided.

During evaluation stability data of only one month and 48 months of only were provided. The CHMP considered that as per ICH Q1A (R2) the long-term stability testing of a new finished product should cover a minimum of 12 months' duration on at least three primary batches at the time of submission, therefore, the data provided was not sufficient to derive a shelf-life for the product. Therefore, the CHMP raised as MO that further stability data should be provided, and shelf life should be proposed

based on the updated data. In addition, results of dissolution in the stability data should be presented for each tested capsule separately. Moreover, photostability testing results were missing for the finished product and should be conducted on at least one primary batch of the drug according to the standard conditions for photostability testing as described in ICH Q1B. In the response, 12 month stability data were provided as requested, therefore, the MO was considered resolved.

Samples were tested for assay, related substance, water content, and dissolution. The analytical procedures used are stability indicating.

One batch was exposed to light as defined in the ICH Guideline on Photostability Testing of New Drug Substances and Products. The results show that the finished product is stable under light stress conditions. Therefore, there is no need for a storage label regarding light.

Two sets of forced degradation studies (thermal, thermal-humidity, photolytic and oxidation) were performed, one to confirm that the assay method was stability indicating and one to confirm the related substance method was stability indicating. These studies were done on the 20 mg capsules which is considered representative of the 10 mg strength. Overall, the forced degradation studies demonstrate the assay method and related substance method are stability indicating and that the finished product is very chemically stable

Based on available stability data, the proposed shelf-life of 24 months without special storage conditions is acceptable.

Adventitious agents

Gelatine obtained from bovine sources is used in the product. Valid TSE CEP from the suppliers of the gelatine used in the manufacture is provided.

2.4.4. Discussion on chemical, pharmaceutical and biological aspects

Information on development, manufacture and control of the active substance and finished product has been presented.

During the procedure, two MOs were raised by the CHMP related to (1) risk assessment and need for confirmatory testing of nitrosamines and (2) stability of the finished product. In relation to nitrosamines, the confirmatory testing is considered necessary regarding nitrosamines originating from the impurity ANA001-31 (mono-methyl derivative of the active substance) and dimethylamine, therefore, the CHMP requested to provide the confirmatory testing as MO and this issue is considered outstanding. As a response to the second MO, 12 month stability data in three commercial scale batches and one batch photostability data were provided, this was considered satisfactory and the MO was resolved.

Moreover, to be in line with EMA Guideline on process validation for finished products (EMA/CHMP/CVMP/QWP/BWP/70278/2012-Rev1, Corr.1) either a detailed process validation plan) or process validation data of the validation batches should be provided and updated sections 3.2.P.5.2 and 3.2.P.5.3. were requested to the applicant and they are still pending.

2.4.5. Conclusions on the chemical, pharmaceutical and biological aspects

In conclusion, one major issue has been identified for the finished product regarding potential presence of nitrosamines. Moreover, other concerns raised by the CHMP for the finished product need to be adequately resolved before the marketing authorisation can be granted. At this stage of the procedure

the CHMP concluded that the quality dossier for the medicinal product Blarcamesine Anavex cannot be recommended for approval.

2.4.6. Recommendation(s) for future quality development

Not applicable.

2.5. Non-clinical aspects

2.5.1. Introduction

Blarcamesine (also termed ANAVEX2-73) is a small molecule, aminotetrahydrofuran derivative, which has been developed as agonist of the Sigma 1 receptor (S1R) and as modulator of muscarinic acetylcholine receptors (M1 to M5).

2.5.2. Pharmacology

2.5.2.1. Primary pharmacodynamic studies

Blarcamesine (ANAVEX2-73) and its primary active metabolite ANAVEX19-144 share a single chiral centre and, hence, are racemates of R- and S-enantiomers. Both parent compound and metabolite revealed high agonist affinity for S1R compared to S2R receptors at small micromolar concentrations of 0.71 and 0.89 µM, respectively, and a 0.2- to 3.5-fold lower affinity at muscarinic M1 to M5 receptors. The affinities of the respective enantiomers of blarcamesine and ANAVEX19-144 were comparable to the data obtained for the racemates.

The pharmacodynamic *in vivo* activity of blarcamesine was investigated following induction of dementia-like AD symptoms by either intracerebroventricular injections of Aβ25–35 fragments (Aβ25–35 mice), the non-selective muscarinic receptor antagonist scopolamine or the non-competitive NMDA receptor antagonist dizocilpine and in transgenic mice containing a double mutation of the human APP gene (Tg2576 mice). Specific agonists and antagonists were included to corroborate the mechanistic activity of blarcamesine.

In Aβ25–35 mice, ≥0.1 mg/kg i.p. blarcamesine dose-dependently reduced the hyperphosphorylation of tau, the seeding of Aβ and of the C99 cleavage product of APP as well as the lipid peroxidation in the hippocampus (Lahmy *et al*., 2013 and 2015). The reduction of lipid peroxidation as a measure of oxidative stress was abolished by concomitant administration of the selective S1R antagonist BD1047 (Lahmy *et al*., 2015). The favourable reductions of these parameters were accompanied by improved memory function in mice treated with blarcamesine, the specific S1R agonist PRE-084 or the muscarinic agonist xanomeline (Lahmy *et al*., 2013).

In line with these findings, blarcamesine doses of 3 mg/kg/day administered via the drinking water for 2 months to transgenic Tg2576 mice improved spatial memory, reduced oxidative stress and increased the expression of synaptic markers in the hippocampus (Lahmy *et al*., 2013a). However, the variable dosing procedure might have precluded the reduction of the Aβ load of the animals by blarcamesine.

Another study in mice revealed the dose-dependent attenuation of learning impairments when i.p. blarcamesine doses of 0.01 - 3 mg/kg were injected 10 min ahead of the induction of dementia-like symptoms by either scopolamine or dizocilpine (Villard V *et al.*, 2011). Doses of 0.3 mg/kg or 1 mg/kg

i.p. blarcamesine or of the ANAVEX19-144 metabolite also reversed the learning deficits of mice when administered 12 weeks prior or one week after injection of aggregated Aβ25–35 peptide fragments. The simultaneous administration of the S1R antagonist BD1047 abrogated the protective effect of blarcamesine in mice treated with dizocilpine or aggregated Aβ25–35 peptide fragments (Villard V *et al.*, 2011).

Blarcamesine was also found to concentration-dependently enhance autophagy up to 2-fold in immortalised human cells *in vitro* or mutant nematodes *in vivo* (Christ *et al.*, 2019). The relevance of these findings was not investigated in the aforementioned mouse models of human AD.

2.5.2.2. Secondary pharmacodynamic studies

In radioligand binding assays, blarcamesine revealed lower affinities of 2.1 to 6.3 µM for the sodium channel, opioid, serotonin and NMDA receptors than for S1R and muscarinic cholinergic receptors. The metabolite ANAVEX19-144 also showed low affinities of 6.6 to 10 µM for the dopamine transporter, sodium channel and κ-opioid receptor, but demonstrated clear interaction with the µ-opioid receptor (0.98 µM) and the NMDA receptor (0.69 - 0.76 µM).

Further pharmacodynamic *in vivo* data in animal models of Parkinson disease, Multiple Sclerosis, Rett syndrome, Fragile X syndrome and different types of seizures indicate that the proposed protective activity of blarcamesine via S1R and muscarinic receptors is not specific for AD and dementia but is generally applicable to various neurological and neurodegenerative conditions.

2.5.2.3. Safety pharmacology programme

The safety pharmacological effects of blarcamesine on vital functions were investigated in core battery studies in accordance with ICH S7A and ICH S7B requirements (CPMP/ICH/539/00 and CPMP/ICH/423/02). The IC_{50} of blarcamesine for inhibition of hERG currents *in vitro* was 26 µM, which translates into a more than 1000-fold safety margin, if the C_{max} of blarcamesine in humans receiving the maximum recommended dose (MRHD) of 40 mg/day and the protein-binding of 71.8 % is considered. Accordingly, no changes in ECG parameters were apparent during cardiovascular evaluation of telemetered Beagle dogs.

Blarcamesine impaired respiratory function within 4 h after the highest single oral dose of 30 mg/kg in a plethysmography study in conscious rats. The significant increases in respiratory rate and airway resistance as well as significant decreases of inspiration time, expiration time and tidal volume indicated the induction of respiratory depression and bronchoconstriction.

Single oral doses ≥10 mg/kg blarcamesine also demonstrated signs of CNS stimulation within 4 h post dose in a standardised functional observation battery in rats as evident by significant numbers of rats moving around the cage. In addition, significantly increased awareness, response to finger approach or head touch, rearing (each within 6 h post dose), stereotypic sniffing, positional passivity, fear response and spontaneous locomotor activity (each within 4 h post dose) were eminent. The NOEL was 3 mg/kg.

2.5.2.4. Pharmacodynamic drug interactions

The combinatorial administration of 0.1 mg/kg i.p. blarcamesine with 0.25 mg/kg i.p. of the acetylcholinesterase inhibitor donepezil improved the behavioural deficits in the Aβ25–35 mouse model of human AD, while the combination with the NMDA receptor antagonist memantine was ineffective (Maurice, 2016).

2.5.3. Pharmacokinetics

The pharmacokinetic properties of blarcamesine were investigated *in vitro* and after single and repeated administration in animals *in vivo*.

Blarcamesine readily permeated Caco-2 cellular monolayers in both apical to basal and basal to apical directions *in vitro*. Accordingly, blarcamesine demonstrated comparably rapid absorption in male mice after single oral intravenous or intraperitoneal administrations. The oral t_{max} of blarcamesine and its primary active metabolite ANAVEX19-144 was 0.5 h. The C_{max} and AUC of blarcamesine and ANAVEX19-144 increased dose-proportionally. The plasma half-lives ranged from 0.6 – 2.3 h for blarcamesine and 2 – 4.6 h for ANAVEX19-144. The absolute oral bioavailability of blarcamesine was 4.43 %, whereas the oral bioavailability of ANAVEX19-144 relative to i.v. administration was clearly higher and amounted to 49.8 %.

The absorption determined after single oral doses in adult and juvenile PND21 rats are consistent with the pharmacokinetic characteristics in mice. However, the t_{max} was delayed in PND21 rats ranging from 0.5 – 2 h for blarcamesine and from 1 – 8 h for ANAVEX19-144.

Following multiple oral administrations of blarcamesine in repeat-dose toxicity studies, the more than dose-proportionally increased exposure of blarcamesine and ANAVEX19-144 was confirmed in adult, juvenile and pregnant rats as well as in dogs. In adult non-pregnant and pregnant rats, both blarcamesine and ANAVEX19-144 accumulated after prolonged dosing for 28 days and longer. In both adult and juvenile rats, the blarcamesine exposure was notably higher in females than in males, but this sex-difference was less pronounced for ANAVEX19-144.

No clear difference between male and female dogs was apparent. The blarcamesine exposure decreased during repeated oral dosing in dogs and was considerably lower upon termination of the respective studies compared to first dose levels. In contrast, the ANAVEX19-144 exposure was substantially higher in dogs than in rats receiving the same blarcamesine doses. In addition, ANAVEX19-144 tended to accumulate following high doses ≥15 mg/kg in dogs.

Blarcamesine showed moderate *in vitro* plasma protein binding ranging from 61.4 % to 71.6 % in animals compared to 71.8 % in humans. Radioactively labelled blarcamesine widely distributed into tissues of pigmented and albino rats within 1 h post dose followed by rapid elimination by 48 – 72 h thereafter. The highest amounts of radioactivity were detected in endocrine (adrenal, pituitary and thyroid glands), metabolic/excretory (liver, bile, kidney and urinary bladder), and secretory tissues (pancreas, lachrymal, Harderian and salivary glands), as well as the gastrointestinal tract (oral mucosa, oesophagus, stomach, cecum and intestine). Contrary to these tissues, clearly lower levels of radioactivity were determined in brain and spinal cord of both rat strains with just up to 2-fold tissue/plasma ratio. The CNS radioactivity disappeared by 8 h post dose in pigmented rats, whereas traces remained detectable by 24 h in albino rats. However, a significant amount of radioactivity persisted in the uveal tract of pigmented rats until termination at 31 days post dose suggesting possible binding to melanin-containing eye structures.

The transfer of blarcamesine or of ANAVEX19-144 across the placenta and into milk was not investigated.

Different proportions of the original blarcamesine concentration were rapidly metabolised within 30 min in liver microsomes of rats (78 %), dogs (54 %) and humans (39 %). The different metabolic rates in the three species are also reflected by the conversion of 57 %, 43 % and 25 % of the initial blarcamesine concentration into the prominent demethylated ANAVEX19-144 metabolite. Profiling experiments in human liver microsomes or with recombinant human CYP isozymes *in vitro* indicate that CYP3A4/5, CYP2C19 and CYP2B6 are involved in the metabolism of blarcamesine, whereas ANAVEX19-

144 is transformed by CYP3A4/5, CYP1A2 and CYP2C8. Further characterisations of the metabolic pathway of blarcamesine were not performed.

Mass-balance evaluations in intact and bile duct-cannulated rats revealed that blarcamesine-related radioactivity was predominantly eliminated within 48 h post oral dose. While roughly equal amounts of radioactivity were excreted by urine and faeces in intact male rats, about 2-fold higher radioactivity counts were determined in urine compared to faeces of intact females. In bile duct-cannulated rats, the proportion of radioactivity was >5-fold higher in bile than in urine of males, whereas this was less pronounced in females with just 1.4-fold higher levels in bile than in urine. Thus, blarcamesine is subject to enterohepatic recirculation with overt sex differences in the metabolism and elimination in rats.

Blarcamesine and ANAVEX19-144 did not demonstrate clinically relevant direct or time-dependent inhibition of CYP isozymes when analysed at sufficiently high concentrations in accordance with ICH M12 recommendations (EMA/CHMP/ICH/652460/2022). Moreover, blarcamesine did not show clinically relevant induction of CYP1A2, CYP2B6 and CYP3A4 mRNA and enzymatic activities in human hepatocytes.

2.5.4. Toxicology

2.5.4.1. Single dose toxicity

Single-dose toxicity of blarcamesine in mice and rats with oral doses of 125 to 1000 mg/kg or i.v. injection from 125 to 250 mg/kg revealed highly similar findings. Clinical signs before death at ≥250 mg/kg mice and ≥500 mg/kg rats involved lack of spontaneous motor activity as well as functional and neurobehavioural deficits (passivity, fear, motor incoordination and impaired reflexes, startle response, muscle tone). In addition, twitches and tremors were observed. Consequently, the maximum tolerated dose (MTD) of orally administered blarcamesine was 125 mg/kg in mice and 250 mg/kg in rats.

Intravenously injected blarcamesine was less tolerated leading to mortality shortly after doses ≥65 mg/kg in mice and rats. The i.v. MTD was 30 mg/kg in both species.

2.5.4.2. Repeat dose toxicity

Repeat-dose toxicity studies with oral administration of blarcamesine for up to 6 months in rats and 9 months in dogs in compliance with GLP and ICH M3(R2) requirements (EMA/CPMP/ICH/286/1995) particularly showed prominent and highly consistent dose-dependent neurological impairments in both animal species.

Oral blarcamesine doses ≥30 mg/kg/day increased the spontaneous locomotor activity in rats shortly after administration or at lower doses on day 1, which then decreased on days 3 and 4. Higher doses ≥45 mg/kg/day were associated with motor incoordination (abnormal gait), reduced spontaneous activity, clonic seizures and piloerection. At ≥60 mg/kg/day, stereotyped movements, twitches, respiratory impairment (dyspnoea), reduced muscle tone (grip strength, body tone), awareness (visual pacing), recumbent position/lying on side, ptosis, chromodacryorrhoea and hollow flanks were noticed, while higher short-term doses of 125 mg/kg/day caused cyanosis and catatonia. Ultimately, the deaths in rats were accompanied by severe clinical signs and appeared to be independent of the treatment duration.

In individual dogs, lower doses ≥5 mg/kg/day blarcamesine increased salivation, reduced spontaneous

locomotor activity and induced tremors, vomiting or liquid faeces. These clinical signs manifested within one to three days after administration of ≥15 mg/kg/day blarcamesine and further aggravated in severity at ≥30 mg/kg/day including tremors, seizures, ataxia, stereotypic movements, a low abdominal tone, the lack of fear and motor coordination, flattened position/lying on side and dyspnoea. Motor incoordination was mainly evident within the first week of repeated dosing in dogs, while it was generally preserved throughout the treatment periods in rats.

Macroscopic examinations at necropsy revealed dose-dependently increased liver weights in rats and dogs treated with ≥15 mg/kg/day blarcamesine (up to +32 % and +41 %, respectively) and higher adrenal gland weights (up to +42 %) in rats. Histologically, these changes corresponded to centrilobular hepatocellular hypertrophy and midzonal vacuolisation as well as basophilia in the adrenal *zona glomerulosa* and minimal vacuolisation of the adrenal *zona fasciculata*. The liver hypertrophy was associated with increased cholesterol, triglycerides, hepatic enzymes (alkaline phosphatase, aminotransferases) and/or decreased total bilirubin at blarcamesine doses ≥5 mg/kg/day in rats and ≥2.5 mg/kg/day dogs, respectively. In addition, dose-dependent thymus atrophy was identified at blarcamesine doses ≥15 mg/kg/day in rats.

2.5.4.3. Genotoxicity

Standard battery genotoxicity studies of blarcamesine were conducted in compliance with ICH S2(R1) and GLP requirements. Blarcamesine was negative for mutations in a reverse mutation assay up to the highest concentrations of 1500/3000 µg/plate with and without metabolic activation in bacteria.

During metaphase analysis in human peripheral blood lymphocytes *in vitro*, blarcamesine was negative for chromosomal aberrations and considered not clastogenic. Nevertheless, blarcamesine showed a possible aneugenic potential by the induction of numerical aberrations at ≥0.93 mM with metabolic activation and at ≥0.58 mM without metabolic activation at acceptable cytotoxic levels. However, a subsequent micronucleus assay in human peripheral blood lymphocytes was negative for chromosomal aberrations, polyploidy, and endoreduplication up to the highest non-cytotoxic concentrations of 2 mM). The slight significant increase in mononucleated lymphocytes observed at 0.25 mM with metabolic activation was not dose dependent and therefore not considered relevant.

Accordingly, blarcamesine did not increase micronucleated PCEs in a micronucleus study up to the highest dose of 50 mg/kg in rats. Clinical signs were comparable to those observed in general toxicology studies in rats. The bone marrow exposure of blarcamesine was not confirmed but can be assumed based on distribution data in rats. The plasma levels of blarcamesine in rats translate into 7 - 19.2-fold safety margins regarding clinical peak plasma levels. Thus, the aneugenic potential observed in the metaphase analysis *in vitro* could neither be confirmed in human peripheral blood lymphocytes *in vitro*, nor in the *in vivo* micronucleus test in rats.

2.5.4.4. Carcinogenicity

Carcinogenicity studies were not conducted, and the proposal of the applicant to perform those after a potential approval is insufficiently justified.

2.5.4.5. Reproductive and developmental toxicity

The developmental and reproductive toxicity (DART) of blarcamesine was investigated in a limited program of studies on fertility and early embryonic development (FEED) in male and female rats, embryo-foetal development (EFD) in rats and rabbits as well as juvenile toxicity studies in rats. The pivotal EFD studies in rats and rabbits, as well as those in juvenile rats, were supplemented by

toxicokinetic measurements. Potential transfer of blarcamesine and its metabolite Anavex-19-144 across the placenta and potential excretion into breast milk were not investigated.

The FEED study revealed reduced oestrous cycling at ≥2.5 mg/kg/day blarcamesine and increased average numbers of early resorptions and post-implantation losses at 15 mg/kg/day in female rats. Additionally, the food consumption and body weight were lowered in male and female rats of this high dose group. It was not reported whether these effects were reversible. However, there was no impact on mating and fertility. A NOAEL of for early embryonic development and female and male fertility was reported.

The pivotal EFD toxicity study in rats revealed blarcamesine-related visceral und numerous skeletal abnormalities in foetuses. While at a test item-related effect with regard to skeletal abnormalities in foetuses/litters was clearly observed, at 5 mg/kg/day this cannot be ruled out beyond reasonable doubt. Hence, the proposed NOAEL of 5 mg/kg/day is not supported.

The pivotal EFD study in pregnant rabbits did not show any impact of blarcamesine on foetal development. The lack of meaningful toxicokinetic exposure data questions the appropriateness of the study design or its implementation and the reliability of the results.

The two studies, which investigated potential adverse effects of blarcamesine in juvenile male and female rats, are of limited relevance for the current clinical indication in adult patients and have therefore not been commented or assessed in detail.

2.5.4.6. Toxicokinetic data

Toxicokinetic determinations showed the more than dose-proportional exposure of blarcamesine and ANAVEX19-144 in plasma samples of adult, pregnant and juvenile rats as well as in dogs. The blarcamesine exposure was generally higher in adult female compared to male rats. This sex difference is reflected in the different excretion profiles but was neither found in juvenile rats, nor in dogs. Adult and pregnant rats also revealed accumulation of blarcamesine and ANAVEX19-144, which was not confirmed in juvenile rats. In dogs, only ANAVEX19-144 tended to accumulate, whereas the blarcamesine exposure decreased upon prolonged dosing. Toxicokinetic determinations in pregnant rabbits indicated the insufficient exposure of the animals.

2.5.4.7. Local Tolerance

No local tolerance studies have been conducted in accordance with the pertinent European guideline (EMA/CHMP/SWP/2145/2000 Rev. 1, Corr. 1*).

2.5.4.8. Other toxicity studies

The primary metabolite ANAVEX19-144 was investigated in single oral dose toxicity studies in mice and rats and after repeated dosing for 7 days in rats. In non-GLP compliant single dose toxicity studies, the lowest oral dose of 62.5 mg/kg ANAVEX19-144 induced hyperreactivity in mice and dose-dependently increased neurological impairments including tremors and clonic seizures at higher levels. The MTD was 250 mg/kg.

Neurological impairments were also detected following single oral doses ≥40 mg/kg ANAVEX19-144 in a GLP compliant study in rats (reduced spontaneous activity, slow and stereotype movements, ataxia, hypotonic gait, prone position, muscle twitches and low body temperature). Repeated oral doses of 40 mg/kg/day ANAVEX19-144 for 7 days confirmed the neurological deficiencies in another GLP compliant study rats.

Two GLP compliant mechanistic neurotoxicity studies in rats also corroborated the neurological impairments after single oral blarcamesine doses ≥30 mg/kg. Both investigations further suggested that single oral doses of 45 mg/kg blarcamesine or above may induce neurodegeneration over time, because neuronal necrosis was detected in the retrosplenial cortex at 72 h post dose but not at the earlier 6 h time point.

2.5.5. Ecotoxicity/environmental risk assessment

A Phase I environmental risk assessment of blarcamesine hydrochloride in accordance with the pertinent European guideline (CHMP/SWP/4447/00 Rev.1) indicates that the PECsurfacewater value of 25 ng/l exceeds the threshold of 10 ng/l. Consequently, a complete Phase II environmental risk assessment is required, which should include an OECD 305 compliant study. The necessary studies would have to be submitted as a post-authorisation measure by Q1 2027, preferably via Type II variation. The applicant provided a signed letter of agreement to this measure with an anticipated timeline.

The applicant provided a study report for an experimental log Dow-study which is accepted. Since the log Dow values of 0.2, 1.7 and 3.3 (for pH 5, 7 and 9, respectively) are below the trigger value, a definitive PBT/vPvB assessment is not needed.

Table 1. Summary of main study results

Substance (INN/Invented Name): Blarcamesine hydrochloride			
CAS-number (if available): 195615-84-0			
PBT screening		Result	**Conclusion**
Bioaccumulation potential- log K_{ow}	OECD107 Study not included	0.2 (pH = 5) 1.7 (pH = 7) 3.3 (pH = 9)	Potential PBT: N open
Phase I			
Calculation	**Value**	**Unit**	**Conclusion**
PEC_{sw}, default	0.25	µg/L	≥0.01 threshold: Y
Other concerns (e.g. chemical class)			

2.5.6. Discussion on non-clinical aspects

Primary pharmacodynamics

Blarcamesine and its active metabolite ANAVEX19-144 demonstrated higher agonist affinity for S1R than S2R receptors at small micromolar concentrations and a lower affinity to muscarinic M1 - M5 receptors in two *in vitro* investigations. In addition, ANAVEX19-144 revealed high affinity for the NMDA and the µ-opioid receptors within the range of S1R and muscarinic cholinergic interactions and blarcamesine dose-dependently reduced dementia-like learning impairments which had been induced by the NMDA receptor antagonist dizocilpine in mice (Villard V *et al.*, 2011). Thus, a potential pharmacodynamic action of blarcamesine and ANAVEX19-144 on the NMDA receptor cannot be ruled out. It should be also noted that the presumptive µ-opioid receptor interaction matches the neurological deficiencies which were consistently observed in toxicity studies in mice, rats and dogs These deficiencies coincide with commonly observed clinical adverse reactions of dizziness, agitation, balance disorders/fall, somnolence, lethargy, fatigue and confusion in human patients. Therefore, further characterisation of the likely action of blarcamesine and ANAVEX19-144 on the NMDA and µ-opioid receptors as well as other potential targets is warranted in a comprehensive secondary pharmacodynamic interaction study *in vitro*.

The pharmacodynamic *in vivo* activity of blarcamesine was investigated in established mouse models

of human AD (Chavan *et al.*, 2023), i.e. following induction of dementia-like AD symptoms by either intracerebroventricular injections of Aβ25–35 fragments (Aβ25–35 mice), the non-selective muscarinic receptor antagonist scopolamine or the non-competitive NMDA receptor antagonist dizocilpine and in transgenic mice containing a double mutation of the human APP gene (Tg2576 mice). Specific agonists and antagonists were included to corroborate the mechanistic activity of blarcamesine.

Blarcamesine doses ≥0.3 mg/kg i.p. consistently prevented the memory deficits and learning impairments in different mouse models of AD and dementia, which was characterised by reduced Aβ seeding, tau hyperphosphorylation and oxidative stress. The parallel testing of other agonists for S1R (PRE-084) and muscarinic receptors (xanomeline) or of the respective antagonists for S1R (BD1047) and muscarinic receptors (scopolamine) suggests, that the pharmacodynamic effects of blarcamesine are mediated via S1R and muscarinic receptors. However, the relative contribution of the S1R agonistic and muscarinic modulatory activity of blarcamesine were not unravelled and scientists regarded blarcamesine as a mixed S1R and muscarinic receptor agonist (Villard V *et al.*, 2011; Lahmy V *et al*., 2013). The applicant should have provided sufficient data to support the claimed mechanistic activities in the product information.

Apart from the known limitations of the tested mouse models of AD, the administration of blarcamesine by the i.p. route or via drinking water likely altered its distribution in mice compared to the intended oral therapy in AD patients. Despite these shortcomings, however, the non-clinical *"proof-of-concept"* is regarded sufficient to support the clinical development of blarcamesine in AD and dementia, considering also the unavailability of more reliable animal models in this indication. Nonetheless, the efficacy of blarcamesine including the identification of an effective dose range need to be demonstrated in human AD and dementia patients (see clinical evaluation).

Secondary pharmacodynamics

Possible off-target interactions were insufficiently investigated for blarcamesine and only included a small panel of presumptive targets in radioligand binding assays. The results of study nos. ANAVEX2-73B05 and ANAVEX2-73B14 were ignored and should be presented.

It seems noteworthy that the primary active metabolite ANAVEX19-144 consistently demonstrated high affinity for the μ-opioid receptor (0.98 μM) and the NMDA receptor (0.69 - 0.76 μM) within the range of S1R (0.89 μM) and muscarinic cholinergic interactions (0.37 – 3.1 μM), which may contribute to the proposed pharmacological activity, explain the observed adverse CNS-related effects in animals and human patients and/or potentially lead to dependence, respectively. As blarcamesine is claimed to exert a novel mechanism of action without licensed predecessor and the affinity of blarcamesine and ANAVEX19-144 to additional targets remains unknown, particularly regarding the CNS, the applicant should have provided a more comprehensive receptor interaction *in vitro* study to define adequate risk minimisation measures for marketing authorisation.

Safety pharmacology

Core battery safety pharmacology studies were conducted in compliance with GLP, ICH S7A and ICH S7B requirements (CPMP/ICH/539/00 and CPMP/ICH/423/02). The IC_{50} of blarcamesine for inhibition of hERG currents *in vitro* was 26 μM, which translates into a more than 1000-fold safety margin, if the C_{max} of blarcamesine in humans receiving the maximum recommended dose (MRHD) of 40 mg/day and the protein-binding of 71.8 % is considered. Except cases of tachycardia at a single time point of the 28 days repeat-dose toxicity study in dogs, ECG recordings remained within normal limits. Likewise, cardiac function was not significantly impaired in human patients. Hence, no arrhythmogenic risk is anticipated for blarcamesine (see clinical assessment).

Blarcamesine induced signs of respiratory depression and bronchoconstriction within 4 h after a single oral dose of 30 mg/kg in a plethysmography study in conscious rats, which coincides with impaired

respiratory function at this or higher dosages in repeat-dose toxicity studies in rats and dogs and is attributable to the prominent CNS-depressant effect of blarcamesine (see below).

In a standardised functional observation battery in rats, single oral doses ≥10 mg/kg blarcamesine also demonstrated signs of CNS stimulation. CNS stimulatory effects like dizziness, confusion, agitation and balance disorders were commonly noted in the clinical program.

Pharmacodynamic drug interactions

Published results of the combinatorial administration of 0.1 mg/kg i.p. blarcamesine and 0.25 mg/kg i.p. of the acetylcholinesterase inhibitor donepezil indicate improved behavioural deficits in Aβ25–35 mice (Maurice, 2016). Of note, the co-administration of blarcamesine with anticholinergic medicinal products may increase clinical side effects.

Pharmacokinetics

Blarcamesine and its active metabolite ANAVEX19-144 were quantified by validated liquid chromatography with tandem mass spectrometry (LC-MS/MS) in plasma of rats, rabbits and dogs. The repeatability and reproducibility of this analytical method was adequately confirmed through incurred sample reanalysis.

Single and repeated oral administrations of blarcamesine revealed fast and more than dose-proportional absorption in rats and dogs. Radioactively labelled blarcamesine widely distributed into tissues. Highest amounts were readily determined in endocrine (adrenal, pituitary and thyroid glands), metabolic/excretory (liver, bile, kidney and urinary bladder), and secretory tissues (pancreas, lachrymal, Harderian and salivary glands), as well as the gastrointestinal tract (oral mucosa, oesophagus, stomach, cecum and intestine). While the distribution into the CNS (brain and spinal cord) was fast (1 h), the radioactivity then successively disappeared by 8 h. In addition, a significant amount of radioactivity persisted in the uveal tract of pigmented rats until termination at 31 days post dose suggesting possible binding to melanin-containing eye structures. Despite the absence of overt ocular toxicity in animals and human patients, the phototoxic potential of blarcamesine has not been adequately addressed.

Blarcamesine was rapidly metabolised at distinct metabolic rates in liver microsomes of rats (78 %), dogs (54 %) and humans (39 %), which is also reflected in the different metabolic conversions into ANAVEX19-144 of 57 %, 43 % and 25 % of the initial blarcamesine concentration. Following prolonged dosing of blarcamesine in rats blarcamesine and ANVEX19-144 accumulated with higher blarcamesine exposure in females than in males, but this sex difference was less obvious for ANAVEX19-144. In contrast, repeated administrations in dogs resulted in comparably decreased blarcamesine exposure in males and females, whereas ANAVEX19-144 tended to accumulate leading to higher exposures in dogs than in rats at the same blarcamesine dose levels. This indicates species-specific metabolic differences between rats and dogs.

Profiling experiments in human liver microsomes or with recombinant human CYP isozymes indicated that blarcamesine was metabolised by CYP3A4/5, CYP2C19 and CYP2B6, whereas ANAVEX19-144 was transformed by CYP3A4/5, CYP1A2 and CYP2C8. However, the applicant did not further characterise the metabolic pathway of blarcamesine *in vitro* and *in vivo*. Accordingly, the earlier request of the pre-submission meeting to compare metabolic profiles between animals and humans was not followed. It is unknown from the mass-balance evaluation in rats, if blarcamesine and/or ANAVEX19-144 are excreted unchanged, because only radioactivity counts were determined in the excreta. Nevertheless, the rapid elimination within 48 h post oral dose was roughly equal in urine and faeces of intact male rats, whereas the renal excretion was 2-fold higher in intact females. Moreover, the pronounced 5-fold higher biliary compared to renal elimination in bile duct-cannulated male rats suggests enterohepatic recirculation, which was minor in females given the just 1.4-fold elevated excretion in bile compared to

urine. Thus, the exposure differences in male and female rats correlate with overt sex-specific metabolism and elimination. Metabolic rates and exposures were also divergent between rats and dogs. A human mass-balance study is not available to elucidate the clinical relevance. It is likely that other phase 1 and/or phase 2 enzymes are involved and the chiral centre within the tetrahydrofuran ring of blarcamesine and ANAVEX19-144 even renders stereoselective metabolism possible. Consequently, ANAVEX19-144 may not constitute the only major metabolite across species including humans. In order to determine the predictive value of the toxicity program of blarcamesine in terms of human safety, the applicant has not sufficiently clarified presumptive qualitative and/or quantitative differences in the metabolic profile between animals and humans.

These metabolic and elimination characteristics corroborate the need for a thorough evaluation of potential pharmacokinetic interactions. Blarcamesine and ANAVEX19-144 did not show clinically relevant direct or time-dependent inhibition of CYP isozymes or induction of CYP1A2, CYP2B6 and CYP3A4 mRNA and enzymatic activities in human hepatocytes when analysed in accordance with ICH M12 recommendations (EMA/CHMP/ICH/652460/2022).

Of note, the efflux ratio of blarcamesine was slightly reduced in Caco-2 cells in the presence of a dual P-gp and BCRP inhibitor indicating a contribution of both transporters. However, potential interactions with other hepatic or renal uptake/efflux transporters in accordance with ICH M12 requirements were neither evaluated for blarcamesine nor for the more stable active metabolite ANAVEX19-144. This deficiency is severe given the unknown metabolic pathway of blarcamesine, the absence of any mass-balance evaluation in humans and the involvement of both urinary and faecal routes in the elimination of blarcamesine-related radioactivity in the mass-balance study in rats. The potential induction of CYP enzymes by ANAVEX19-144 has also not been tested. As most AD patients are of advanced age with possibly compromised hepatic and renal clearance and will likely require co-medications, the adequate characterisation of pharmacokinetic drug interactions is mandatory and should have been available already for early drug development. The applicant did not adequately respond to this deficiency, so the absence of these data therefore continues to constitute a major objection against marketing authorisation of blarcamesine.

Toxicology

The toxicology of blarcamesine was investigated in compliance with GLP and ICH M3(R2) requirements (EMA/CPMP/ICH/286/1995) by the oral route intended for clinical therapy in single and repeat-dose studies for up to 26 weeks in rats and 39 weeks in dogs. In addition, the systemic exposure was maximised by testing single i.v. doses of blarcamesine in mice and rats. The tested blarcamesine batches contained higher impurity levels than the current clinical lot and complied with the present release specification. The species selection has been justified by the general regulatory acceptance for toxicological investigations. However, the missing qualitative and/or quantitative comparison of metabolic profiles between animals and humans limits the predictive value of the toxicity program in terms of human safety.

Single and repeat-dose toxicity

The single and repeat-dose toxicity studies revealed prominent and highly similar dose-dependent neurological impairments in mice, rats and dogs.

In rats, spontaneous locomotor activity was initially increased after blarcamesine doses ≥30 mg/kg/day and then decreased on successive days. Elevated doses ≥45 mg/kg/day blarcamesine reduced spontaneous activity, muscle tone and awareness in rats, while motor incoordination, stereotyped movements, clonic seizures and piloerection were increased.

Blarcamesine doses ≥5 mg/kg/day blarcamesine increased salivation, reduced spontaneous locomotor activity and induced vomiting or liquid faeces in individual dogs, whereas administration of elevated

doses ≥15 mg/kg/day were associated with tremors, seizures, ataxia, motor incoordination, stereotypic movements, low abdominal tone and lack of fear.

The neurological deficiencies in rats and dogs coincide with the commonly reported clinical adverse reactions of dizziness, agitation, balance disorders/fall, somnolence, lethargy, fatigue and confusion in human patients and apparently account for the proposed titration regimen for clinical therapy of blarcamesine. The initial stimulatory effect followed by CNS depression upon prolonged treatment or at higher doses may at least in part be attributable to the presumptive high affinity of the primary blarcamesine metabolite ANAVEX19-144 for the μ-opioid and NMDA receptors.

Macroscopic and microscopic examinations revealed dose-dependently increased liver hypertrophy in association with increased cholesterol, triglycerides, hepatic enzymes and/or decreased total bilirubin in rats and dogs administered ≥15 mg/kg/day blarcamesine. In addition, higher adrenal gland weights with basophilia in the adrenal *zona glomerulosa* and minimal vacuolisation of the adrenal *zona fasciculata* was detected in rats. These liver and adrenal gland abnormalities reversed during the recovery periods and were interpreted as metabolic adaptations. This justification and the NOAEL of is accepted given 1) the liver metabolism of blarcamesine via CYP isozymes involving enterohepatic circulation in rats and 2) scientific knowledge from liver and adrenal hypertrophy following drug treatment (Greaves P, 2012; Hall *et al*., 2012).

Dose-dependent thymus atrophy was detected after ≥15 mg/kg/day blarcamesine in rats, which only partially recovered following higher doses ≥30 mg/kg/day in the 28 days repeat-dose toxicity study. Although the thymus atrophy might reflect the anti-inflammatory activity of blarcamesine as reported for another sigma agonist (Carayon *et al*., 1996), no further discussion on the claimed anti-inflammatory action of blarcamesine was provided.

Genotoxicity

In standard battery genotoxicity studies in compliance with ICH S2(R1) and GLP requirements, blarcamesine was negative for mutations in a reverse mutation assay in bacteria, negative for chromosomal aberrations in human peripheral blood lymphocytes and did not increase micronucleated PCEs in a micronucleus study up to the highest dose of 50 mg/kg in rats. Although the bone marrow exposure of blarcamesine was not confirmed in this study, it can be reasonably assumed based on comparable clinical signs as observed in general toxicology studies and distribution data obtained in rats. Hence, the plasma levels of blarcamesine determined at the same doses in toxicity studies in rats translate into 7 - 19.2-fold safety margins with regard to clinical peak plasma levels. Thus, the aneugenic potential suggested by the metaphase analysis in another *in vitro* study of blarcamesine in human peripheral blood lymphocytes was neither confirmed *in vitro*, nor in the *in vivo* micronucleus study in rats. The genotoxicity of blarcamesine is hence regarded sufficiently addressed and the risk from blarcamesine administration is considered to be low.

Carcinogenicity

No carcinogenicity studies were conducted. Blarcamesine is intended for the chronic treatment of AD and dementia, a severely debilitating condition with very limited treatment options. The applicant provided a very brief and incomplete weight-of-evidence (WoE) approach to justify the postponement of carcinogenicity testing post-approval. Several aspects concerning the carcinogenetic potential were not addressed. There are uncertainties and concerns regarding the novel mode of action, primary and secondary pharmacodynamic, metabolic profiles as well as the insufficient clinical experience with S1R agonists. Although genotoxicity and chronic toxicity results did not raise concerns regarding potential carcinogenicity, possible immunological and endocrine effects were not discussed. To waive a rat carcinogenicity study by a WoE approach, all aspects of the Addendum to the ICH S1B(R1) guideline, Part II) need to be addressed in detail, thoroughly discussed and referenced based on all study results

including, if available, relevant literature data. Thus, it would only have been feasible to postpone carcinogenicity testing post approval had a complete WoE evaluation been submitted in accordance with ICH S1B(R1) requirements, in addition to a signed commitment clarifying that any carcinogenicity studies would be initiated no later than three months post-approval including also a schedule for submission of the final study reports.

Developmental and reproductive toxicology

The presented developmental and reproductive toxicity program of blarcamesine was limited to a FEED study in male and female rats, EFD studies in rats and rabbits and juvenile toxicity studies in rats. In the FEED study, blarcamesine doses of 15 mg/kg/day increased early resorptions and post-implantation losses.

The pivotal EFD study in rats demonstrated visceral und numerous skeletal abnormalities in foetuses in dams administered 5 and 15 mg/kg/day blarcamesine. Hence, a developmental NOAEL of 2.5 mg/kg/day seems to be more appropriate as it also coincides with the results obtained in the preliminary EFD study in rats and entails a recalculation of safety margins with respect to human exposure levels. The pivotal EFD study in pregnant rabbits did not demonstrate any foetal malformations, but lacked meaningful toxicokinetic exposure data, which questions the study design and the reliability of the outcome. However, the unequivocal induction of malformations observed in the two EFD studies with blarcamesine in rats at doses with little or no safety margin compared to the MRHD can be regarded sufficient for the assessment and implementation of risk minimisation measures during pregnancy and further EFD studies are not considered necessary. Nonetheless, changes in bone length and diameter were also noted in juvenile rats.

Of note, no PPND study was conducted and the justification of the applicant for its absence with respect to the envisaged elderly patient population is not endorsed, because the proposed broad clinical indication also includes cases of early dementia, which may already develop in women in their 30s or 40s, i.e. before menopause (Hendriks *et al*., 2021; Li *et al*., 2024). Thus, the use of blarcamesine by women of child-bearing potential and during pregnancy can presently not be excluded. Due to the limited available data indicating, among other issues, the induction of malformation in rats with missing or minor safety margin compared to human exposure, the applicant should have thoroughly elaborated on the need for a contraindication during pregnancy, thereby discussing the presumptive mechanistic basis for the malformations in rats. In addition, the age and sex of the patients in the clinical program of blarcamesine should be clarified. The applicant did not provide this discussion but suggested to contraindicate the administration of blarcamesine during pregnancy and in women of childbearing potential not using effective contraception. Although this contraindication is generally endorsed it is not acceptable without any justification based on a detailed discussion of pertinent non-clinical and clinical data. In addition, the rationale for the suggested contraception duration and the newly proposed contraindication "breastfeeding" should have been substantiated.

Toxicokinetics

The applicant calculated safety margins based on the AUC determined in plasma at the NOAELs in repeat-dose toxicity studies of rats and dogs compared to the estimated median AUC of blarcamesine and ANAVEX19-144 in patients administered a single 30 mg/day dose of blarcamesine. Except the safety margin derived for ANAVEX19-144 in the chronic toxicity study in dogs, all other safety margins are minor, or exposures in animals were even below human levels. It should be noted that lower safety margins can be expected, if the AUC in patients following repeated administrations of the MRHD of 40 mg/day blarcamesine would be available. Consequently, the obvious neurological impairments observed in rats and dogs of the toxicology program correlate with common adverse events in human patients (see sections 4.4 and 4.8 of the proposed SmPC). It is therefore concluded that repeat-dose

toxicity studies in animals do not provide any relevant safety margins with respect to the anticipated exposure of blarcamesine in patients receiving the MRHD.

Dependence

The dependence potential of blarcamesine was not investigated as expected for a new CNS active substance. The applicant questions the scientific rationale regarding the need for dedicated investigations, which grounds on an integrated evaluation of the available non-clinical and clinical data in accordance with the tiered approach of the pertinent European guideline (EMEA/CHMP/SWP/94227/2004).

The claimed mechanistic activity of blarcamesine as agonist of S1R and muscarinic cholinergic receptors seems to be novel and lacks non-clinical and clinical experience from licensed predecessors within the EU or abroad. In addition, blarcamesine can be easily administered via the oral route, was quickly absorbed and distributed into the CNS of rats after 1 h post dose followed by a rapid decline within 8 h. In line with these fast *"on/off"* pharmacokinetic properties, blarcamesine and ANAVEX19-144 initially or at low doses induced CNS stimulation in single and repeat-dose toxicity studies in mice, rats and dogs, which was subsequently followed by salient neurological impairments involving awareness, stereotypic locomotion, motor incoordination, abnormal muscle tone and respiratory depression at elevated dose levels. These clinical findings developed within the first week of treatment and then normalised pointing towards the development of tolerance. They also coincide with clinical adverse reactions.

The primary active metabolite ANAVEX19-144 showed high affinities for the μ-opioid and NMDA receptors in the range of interactions determined for S1R and muscarinic receptors, which could contribute to the observed neurological impairments in animals and human patients. As ANAVEX19-144 is generated by demethylation of blarcamesine, the same interactions can be reasonably assumed for the parent substance and may also apply to further active metabolites. Moreover, further interactions with known targets of dependence can presently not be excluded given the absence of a thorough *in vitro* secondary pharmacodynamic interaction study.

Furthermore, literature data indicate that S1R agonists may potentiate neurotransmitter release including dopamine, cognitive function, act as antidepressant and modulate the activities of cocaine and alcohol (Hayashi and Su, 2004; Maurice and Su, 2009). S1R and NMDA receptors have been also implicated in addictive behaviours, which is well known for the μ-opioid receptor (Contet *et al*., 2004; Hopf, 2017; Aguinaga *et al*., 2019).

The applicant did not provide any new results or literature evidence that could dispel any of these aspects. Although earlier requested and straightforward to perform, the applicant has not extended the investigation of potential secondary pharmacodynamic *in vitro* interactions of blarcamesine and ANAVEX19-144 yet. This study would be a prerequisite for setting adequate risk minimisation measures including the dependence potential and the WoE regarding postponement of carcinogenicity testing. The study should therefore have been completed within this MA procedure. Depending on the outcome of this *in vitro* study, the applicant had been reminded during the procedure to provide a signed commitment for the conduct of behavioural *in vivo* studies testing withdrawal and tolerance, reinforcing properties and drug discrimination phenomena, which would have enabled definition of adequate risk minimisation measures during clinical therapy with blarcamesine.

Metabolites

The primary metabolite ANAVEX19-144 dose-dependently increased neurological impairments in single dose toxicity at ≥62.5 mg/kg in mice and at ≥40 mg/kg in rats, which were highly similar to observations in the 14 days and 28 days repeat-dose toxicity studies of blarcamesine in rats. Neurological deficiencies were confirmed following repeated oral doses of 40 mg/kg/day ANAVEX19-

144 for 7 days in rats. Thus, ANAVEX19-144 mediates at least part of the pharmacological and toxicological effects of blarcamesine.

Phototoxicity

No data or justification for the absence of a phototoxicity evaluation was provided, although this had been explicitly requested in the pre-submission meeting. Blarcamesine was found to distribute into skin and persisted in the uveal tract of the eyes of pigmented rats for more than 30 days suggesting high affinity for melanin. The blarcamesine spectrum submitted in Module 3.2.S.3.1 indicated an absorption maximum in short wave UV light (~200 nm) and a molar extinction coefficient greater than 20,000 $l \cdot mol^{-1} \cdot cm^{-1}$ but was limited to ~490 nm. An extended absorbance spectrum of blarcamesine was subsequently provided, which does not show any relevant absorption in the range from 200 nm to 700 nm but the inconsistency of this new absorption spectrum to that contained in Module 3.2.S.3.1 was not elucidated.

Mechanistic studies

Two GLP compliant mechanistic toxicity studies in rats indicate that single oral doses of ≥45 mg/kg blarcamesine may induce neurodegeneration in the retrosplenial cortex over time. However, the severity was minimal, affected only half of the rats and no neuronal necrosis was identified after an intermediate dose of 60 mg/kg blarcamesine or following repeated blarcamesine administrations of ≥45 mg/kg/day for 28 days in rats. In addition, no neuronal degenerations were observed in other toxicity studies for up to 26 weeks in rats and 39 weeks in dogs. Based on the rat plasma AUC at 45 mg/kg compared to humans administered a once daily clinical dose of 50 mg blarcamesine, the applicant deduced safety margins of about 14-fold for blarcamesine and 6-fold for ANAVEX19-144. Hence, the weak neuronal necrosis in rats is regarded of unlikely clinical relevance.

Environmental risk

As a result of the above considerations, the available data do not allow to conclude definitively on the potential risk of blarcamesine to the environment.

2.5.7. Conclusion on the non-clinical aspects

The non-clinical dossier of blarcamesine is not adequate and harbours the following principal deficiencies, which– together with the other elements leading to a negative benefit-risk (see below), preclude a marketing authorisation:

1. The claimed mechanism of action of blarcamesine as predominant S1R agonist and muscarinic modulator has not been unequivocally corroborated considering the negation of the outcome of several primary pharmacodynamic *in vitro* investigations that also indicate the high affinity of ANAVEX19-144 for the NMDA and the μ-opioid receptors within the range of S1R and muscarinic cholinergic interactions. In order to disentangle the pharmacological activity of blarcamesine and ANAVEX19-144, a comprehensive receptor interaction *in vitro* study is required.
2. The pharmacokinetic program of blarcamesine lacks a qualitative and/or quantitative comparison of metabolic profiles between animals and humans, which limits the predictive value of the toxicity program in terms of human safety. The insufficient investigation of pharmacokinetic interactions precludes a marketing authorisation in an elderly patient population, which frequently has to rely on co-medications.
3. Given the absence of a complete weight-of-evidence evaluation and the lack of a signed commitment, postponement of carcinogenicity testing is not acceptable.

4. Limited developmental and reproductive toxicology studies further indicate that blarcamesine may induce malformations in rats with no or minor safety margins compared to human exposure. As the proposed broad clinical indication includes women of childbearing potential with early-onset dementia in their 30s or 40s, the need for a contraindication during pregnancy and in women of childbearing potential not using effective contraception should have been justified including also the proposed duration of contraception. In addition, the newly suggested contraindication "breastfeeding" should have been substantiated**.**
5. The dependence potential of blarcamesine should be investigated to define adequate risk minimisation measures for the proposed clinical therapy with regard to 1) the presumptive pharmacological profile of blarcamesine and its active metabolite ANAVEX19-144, 2) the easy to administer pharmaceutical form, 3) the fast *"on/off"* pharmacokinetic properties in the CNS and 4) salient and consistent neurological impairments in animals and human patients. Depending on the outcome of the thorough evaluation of secondary pharmacodynamic of blarcamesine and ANAVEX19-144 *in vitro*, a signed commitment for the conduct of behavioural *in vivo* studies testing withdrawal and tolerance, reinforcing properties and drug discrimination phenomena would be required, without which adequate risk minimisation measures cannot be defined.

2.6. Clinical aspects

2.6.1. Introduction

GCP aspects

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

The applicant has provided a statement to the effect that clinical trials were conducted in accordance with all stipulations of the protocol and in accordance with the current International Conference on Harmonisation (ICH) GCP guidelines, the Declaration of Helsinki and local regulations (as applicable).

- **Tabular overview of clinical studies**

Study ID	Enrolment status Start date Total enrolment/ enrolment goal	Design Control type	Study & control drugs Dose, route of administration and duration Regimen	Population Main inclusion/ exclusion criteria
ANAVEX2-73-AD-004 (also referred to as AD-004 or -004), pivotal	Start Date: 27 Aug 2018 (FPFV) Stop Date: 28 Jun 2022 (LPLV) 509 planned; 508 analysed (Placebo N=170, BL 30mg N= 169, BL 50mg N=169)	Phase 2b/3 48-week, double-blind, randomized, placebo-controlled, parallel group	Blarcamesine Target dose of 30 mg or target dose of 50 mg or placebo titrated over a period of 2-3 weeks[a] PO, QD for 48 weeks	Early AD • M or F 60 to 85 years of age • 2011 NIA-AA criteria for diagnosis of MCI due to AD or early-stage mild dementia due to AD, supported by historical records (CSF, PET scan, or CT or MRI scan) • MMSE score 20-28 FCSRT recall score of ≤17 or total score <40
ANAVEX2-73-AD-EP-004, (also referred to as AD-EP-004 or EP-004), pivotal OLE	Start Date: 10 October 2019 (first informed consent Stop Date: 13 June 2024 (last visit) Study terminated by Sponsor on 21 May 2024 The actual number of participants enrolled and analysed was 300.	Phase 2b/3 OLE open-label treatment for 96/144 weeks	Blarcamesine Target dose of 50 mg titrated over a period of 10 weeks[b] PO, QD for 96 weeks (144 weeks in Australia)	Participants who completed the 48-week study, AD-004
ANAVEX2-73-002 (also referred to as AD-002 or -002)	Study Dates: Initiation date – 16 December 2014(First patient first visit) Completion date – 02 November 2016 (Last patient last visit) In Part A, a total of 32 subjects were planned for enrollment. Thirty-two subjects participated in Part A and 30 subjects participated in Part B.	Phase 2a, 2-part study Part A: sequentially randomized, open-label, 2-period crossover of either low or high dose IV and low or high dose oral formulation Part B: open-label, oral treatment for 52 weeks	Blarcamesine IV: 3 or 5 mg PO: 30 or 50 mg with adjustments allowed based on PK and safety Part A: 11 doses in Period 1 and 12 doses in Period 2 over ~35 days Part B: PO, QD for 52 weeks (actual doses were 10, 20, 30, and 50 mg/day)	Mild-to-moderate AD • M or F 55 to 85 years of age • Diagnosis of probable AD in accordance with the DSM-IV and NINCDS-ADRDA criteria • Brain CT or MRI scan within prior 12 months consistent with clinical diagnosis of probable AD • MMSE score 16-28 Rosen Modified Hachinski Ischemic score ≤ 4
ANAVEX2-73-003 (also referred to as AD-003 or -003)	Start Date: 23 Mar 2016 Stop Date: 10 Nov 2020 Up to 32 participants planned 24 participants were enrolled and analysed	An Extension Study of ANAVEX2-73 in Patients with Mild to Moderate Alzheimer's Disease Phase 2 OLE open-label treatment for 208 weeks	Blarcamesine 10, 20, 30, or 50 mg (based on dose at end of study AD-002) PO, QD for 208 weeks	Participants who completed the 52-week Part B in study AD-002

2.6.2. Clinical pharmacology

2.6.2.1. Pharmacokinetics

Absorption, Distribution, Metabolism, Elimination

After oral administration Blarcamesine, also known as ANAVEX2-73, was rapidly absorbed based on a relatively short t_{max} of 1.5 to 2 hours. The metabolite ANAVEX19-144 appeared with a median T_{max} of 2 to 3.5 hours in healthy volunteers and about 6 hours in patients with Alzheimer disease.

There was no dedicated investigation of bioavailability. Oral and IV applications at different doses in patients with AD were compared indirectly through population PK modelling. Absolute bioavailability was estimated to be 27.7% although this calculation is only possible with near-linear PK, which at this time is questionsable. Food effect was not investigated and considered unlikely by the applicant, since blarcamesine is a Class I drug (highly soluble and highly permeable). Low bioavailability might therefore indicate a high first pass effect. Investigation of absolute bioavailability and food effect in dedicated pharmacological studies are requested.

Distribution, metabolism and elimination of blarcamesine in humans are overall not well characterized. The major metabolite (ANAVEX19-144) is formed through cytochrome metabolism in human liver microsomes. Further metabolic pathways were however not characterized either in vitro or in vivo. While the non-clinical characterization of the drug is already considered incomplete no human mass balance study was conducted and the applicant seems to just have assumed that there are no other metabolites present in humans. Renal elimination of blarcamesine was only investigated in the First-In-Human trial where less than 20% of ANAVEX19-144 was found to be eliminated through urine. Therefore, it remains mostly unclear how blarcamesine and its major active metabolite are further metabolised and eliminated. Due to interspecies differences full characterization of elimination and metabolism in humans will require a mass-balance study.

Dose proportionality and time dependencies

There are conflicting data regarding dose proportionality that require clarification. While early single-dose healthy volunteer PK indicated dose proportionality the noncompartmental PK analysis of combined Phase 1 and Phase 2a data indicated less-than-dose proportional kinetics. In contrast, the final combined popPK model indicated greater-than-dose proportional pk.

There has also been no adequate investigation of steady-state PK in either volunteers or patients as there has been only sparse PK sampling of through levels after multiple doses. It is not possible to reliably predict if PK parameters might change over time.

Pharmacokinetics in the target population

In the popPK analysis, a significant difference in the fraction metabolized was found between healthy volunteers and patients. In relation to patients, the fraction of blarcamesine that was metabolized to ANAVEX19-144 was only 23% in healthy volunteers. This difference appears counter-intuitive and the applicant is requested to discuss. The applicant suggests that changes in the gut barrier ("leaky gut") or differences in the microbiome in AD patients may explain the higher levels of the metabolite ANAVEX19-144. However, these factors mainly affect drug absorption in the intestine, not the rate or extent of metabolism in the liver. The FM parameter reflects how much of the parent drug is metabolized into the main metabolite. It is usually related to liver function and enzyme activity, not gut permeability. Therefore, the justification is not sufficient, and the model assumption is still not acceptable.

Special populations

In the popPK analysis neither renal function, hepatic function nor body weight were not detected as a significant covariate on the PK. Race and age were not investigated as covariates.

Pharmacokinetic interaction studies

Only non-clinical interaction studies have been conducted. The metabolic pathways of blarcamesine and not considered fully understood, neither can it be excluded that additional relevant metabolites exist. Therefore, there is a significant risk for undiscovered drug-drug-interaction, which is of grave concern in an elderly population that can be expected to receive significant co-medication.

Evaluation and qualification of popPK model

The popPK analyses show substantial model misspecification for the parent and the metabolite model. Notable issues in the predictive performance across doses and studies are observed. In particular, the study ANAVEX2-73-AD-004 in AD patients is not well described across doses, exposure of lower doses is overpredicted and exposure for higher doses underpredicted. Therefore, the presented model is not considered adequate to describe the pharmacokinetics of blarcamesine. The applicant acknowledged the limitations of the current model and commits to updating the population PK model following the availability of new data. The applicant did not present an update of the model during this procedure.

2.6.2.2. Pharmacodynamics

Mechanism of action

Blarcamesine is an agonist for Sigma-1 Receptor (S1R) and muscarinic receptors. The S1R is a chaperone protein that interacts with diverse client targets including ion channels, endoplasmic reticulum (ER) proteins, neurotrophins, G protein-coupled transporters (GPCRs), ER-localized mRNA translation complexes, and autophagy organelles. It is speculated that S1R mediates and promotes healthy autophagy and drives downstream neuroprotection which improve cognition and behavior. There is no specific action against Alzheimer pathology. It is not possible to measure successful target engagement/activation of Sigma-1 receptor in the CNS.

Primary and Secondary pharmacology

In Study ANAVEX2-73-AD-004 blood levels of AD disease pathology biomarkers including beta-amyloid peptides (Abeta40 and Abeta42), p-Tau (181) and p-Tau (231), Nf-L, YKL-40, BACE1, Glutamate and related metabolites, and other AD disease markers were measured as exploratory efficacy endpoints. Plasma samples were taken at baseline and after 48 weeks.

Aβ42/40 ratio was the only endpoint that showed a statistically significant change compared to placebo. For Nf-L, p-Tau (181), and p-Tau (231) a non-significant numerical superiority was found.

Unspecific CNS side-effects were found as dose limiting toxicity in healthy volunteers for single doses of 60 mg blarcamesine.

Relationship between plasma concentration and effect and safety

Exposure in both Exposure-Response and Exposure-Safety analyses was calculated using the independently developed population PK model. The population PK model displayed substantial model misspecification. Substantial overprediction occurred in the popPK model when predicting lower doses

(10mg) and underprediction occurred with higher doses (50mg). This leads to a flatter dose-exposure relation and less exposure difference between low and high doses. Therefore, while ER analyses might be considered methodologically acceptable, no conclusions can be drawn based on these analyses since the popPK model, which provided the exposure, has substantial limitations.

2.6.3. Discussion on clinical pharmacology

Clinical pharmacology of blarcamesine was assessed in three studies overall. Intensive PK sampling was done in healthy volunteers to characterize pharmacokinetics after single oral doses. Additional intensive PK sampling was done in patients with Alzheimer disease after the first dose only, either given orally or by IV application. No steady state concentration-time profiles are available. Steady state PK data are limited to pre-dose through level measurements in patients.

Multiple dose and steady-state PK was primarily described with a popPK model. The popPK analyses show substantial model misspecification for the parent and the metabolite model. Notable issues in the predictive performance across doses and studies are observed. In particular, the study ANAVEX2-73-AD-004 in AD patients is not well described across doses, exposure of lower doses is overpredicted and exposure for higher doses underpredicted. Therefore, the presented model is not considered adequate to describe the pharmacokinetics of blarcamesine. The Applicant acknowledges the limitations of the current model and commits to updating the population PK model following the availability of new data. A population PK model in a marketing authorisation dossier is not only a descriptive tool. It is also used to understand variability, to identify important covariates, and to predict exposure in subgroups that were not fully studied. A model that needs an extreme, not validated difference in FM between two populations cannot be trusted for such predictions. In summary, the response does not resolve the raised concern. The main issues remain. Thus, the Applicant should have revised the popPK model and provided additional data on the elimination pathways to support the FM assumptions. Without such justification, the model (which was not updated during the procedure) cannot be considered robust for PK prediction.

Bioavailability was not investigated in a dedicated study but calculated indirectly by noncompartmental analysis using data from Phase 1 and 2a studies. The absence of a dedicated food effect study was justified by blarcamesine being a Class I drug (highly soluble and highly permeable). Calculated absolute bioavailability of 27.7% would then indicate a significant first-pass effect, as Class I would indicate rapid and complete absorption. The applicant provided additional explanation for the calculation of bioavailability and the classification as BCS Class I drug. Further, the applicant confirmed, that bioavailability will be further investigated as part of the required mass balance study.

Distribution, metabolism and elimination of blarcamesine in humans have not been sufficiently investigated. Investigation of the metabolism was limited to in-vitro studies of cytochromes. This might address the initial metabolism, additional metabolism involving non-hepatic enzymes is likely but has not been investigated. The applicant states in their responses that CYP2C19 genotype was assessed in study ANAVEX2-73-AD-004 at selected sites. The absence of a formal CYP2C19 polymorphism analysis is not considered a critical deficiency. However, clinical drug–drug interaction studies with strong CYP3A4 and CYP2C19 inhibitors are required to adequately assess the risk of increased exposure and to define appropriate safety measures. Moreover, a dedicated study in patients with hepatic impairment is needed. However, these are considered some of the specific issues in an overall incomplete characterisation of pharmacokinetics.

Less than 20% of the active metabolite ANAVEX19-144 is found in urine. Further elimination of metabolite and parent is unclear and requires additional investigation. The applicant proposed to perform the missing studies as post-authorisation measures under a conditional marketing

authorisation. However, the conditions for a CMA are not met. The mass-balance study in humans, full transporter profiling, and CYP induction studies (for both parent and metabolite) should have been provided before authorisation.

As the metabolism is not fully understood not all relevant enzymes and transporter that are involved in metabolism and elimination pathways are known. It is also possible that additional metabolites have not been properly identified. Consequently, the potential for drug-drug-interaction in humans cannot be considered fully understood also. Given that the target population of Alzheimer patients is elderly and will require co-medication, the risk for drug-drug-interactions is considered significant. The adequate characterization of all involved metabolising enzymes and drug transporters as well as pharmacokinetic drug interactions for blarcamesine, ANAVEX19-144 and any additional major human metabolites is mandatory, and the absence of these data precludes to define adequate risk minimisation measures. The applicant in their response proposed to conduct the requested studies as post-authorisation study after a CMA. This is however not acceptable. The full characterization of the metabolism of blarcamesine and all relevant metabolites must be performed in accordance with ICH M12 requirements before marketing authorisation (including CMA) can be granted.

The active substance blarcamesine hydrochloride contains one chiral centre and is a racemate of equal amounts of R- and S-enantiomers. No data were provided if pharmacological differences between the enantiomers have been investigated and if interconversion happens in-vivo. The applicant has provided IC50 values for the R- and S- enantiomers of ANAVEX2-73 and ANAVEX19-144. As they are overall similar comparable responses should be expected. The question of in-vivo interconversion was not addressed.

There is conflicting information, whether blarcamesine has dose proportional pharmacokinetics. While early single dose data in healthy volunteers showed linear increases of C_{max} and AUC, early modelling based on Phase 1 and Phase 2a data indicated less than dose proportional PK while in the final popPK model PK was found to increase greater than dose proportional. A more detailed discussion and sensitivity analysis would have been expected to explore the possible influence of different mechanisms that may be responsible for the irregularity of Cl/F and T1/2 at higher doses, such as saturable metabolism, changes in transporters, or dose-related changes in bioavailability.

The use of a titration scheme is noted, but approach based on the tolerability does not fully mitigate the need to evaluate accumulation risk, especially in elderly patients with AD. The Applicant should have provided the accumulation indices, time to steady state, and fluctuation metrics for both parent and metabolite, stratified by age and relevant covariates including the patients with disturbances in renal and hepatic function for maximum recommended dose and implications for dosing/titration in elderly.

2.6.4. Conclusions on clinical pharmacology

The assessment of clinical pharmacology in humans is insufficient. Human metabolism and elimination pathways of blarcamesine must be considered only partially known. Also, the non-clinical in-vitro assessment of blarcamesine and active metabolite ANAVEX19-144 was confined to the potential inhibition of CYP isozymes. Interaction with hepatic and renal uptake/efflux transporters in accordance with ICH M12 requirements was assessed for P-gp and BCRP only. Given the unknown metabolic pathway of blarcamesine, the absence of any mass-balance evaluation in humans and the involvement of both urinary and faecal routes in the elimination of blarcamesine-related radioactivity in the mass-balance study in rats these deficiencies are severe. Consequently, pharmacokinetic drug-drug-interaction in humans cannot be considered adequately characterized. This is of great concern, as the target population of AD patients are of advanced age and will likely require co-medication. This and the

absence of a human mass balance study constitutes factors precluding the granting of a marketing authorisation, as the absence of drug-drug-interaction data and insufficient characterisation of elimination and metabolism preclude implementing of adequate risk minimisation measures.

2.6.5. Clinical efficacy

The blarcamesine clinical development program consists of efficacy and safety data from 5 studies involving a total of 886 participants (22 participants in ANAVEX2-73-001, 32 participants in ANAVEX2-73-002, 24 participants in ANAVEX2-73-003, 508 participants in ANAVEX2-73-AD-004 DB, 300 participants in ANAVEX2-73-AD-EP-004). A double-blind, placebo-controlled pivotal 48-week study (Study ANAVEX2-73-AD-004) and its long-term extension up to 144 weeks (Study ANAVEX2-73-AD-EP-004) conducted in patients with early AD and supportive efficacy data from a randomized, open-label 57-week study (Study ANAVEX2-73-002) and its 208-week long-term extension (Study ANAVEX2-73-003) conducted in patients with mild-to-moderate AD constitute the main basis for benefit/risk evaluation.

The clinical development program includes only one pivotal randomised, placebo-controlled study in patients with MCI due to AD or early-stage mild dementia due to AD. There is no replication of the findings, which is especially concerning as blarcamesine has a new mechanism of action. Blarcamesine (ANAVEX2-73) is a Sigma-1 receptor (S1R/SIGMAR1) agonist and to lesser extent muscarinic receptor modulator. The open-label extension ran until May 2024, when it was terminated by the applicant to evaluate the results.

Two additional open label studies have been also submitted, but these have been conducted in mild to moderate AD in a limited number of patients (approximately 30 patients).

2.6.5.1. Dose response study(ies)

No classical dose finding study for efficacy was conducted for blarcamesine. The pivotal study ANAVEX2-73-AD-004 in early AD compared 30mg and 50mg in separate trial arms versus placebo.

ANAVEX2-73-002 was a phase IIa adaptive-trial-design study with repeated doses to find the maximum tolerated dose, investigate pharmacodynamic and bioavailability evaluation in patients with mild to moderate Alzheimer's disease with a 36 days' duration in Part A and a 52-week open label follow-up period in Part B. Two capsule strengths (10 mg and 20 mg) were manufactured to cover the planned oral dose ranges (10 mg to 60 mg). The infusion formulation of the drug substance consisted of.

Dosages of 10 mg, 20 mg, 30 mg 50 mg were studied in the small open label study AVANEX-73-003 (n=26; see section 3.7.2) in a more advanced population with mild to moderate AD. Overall, the dose justification for the doses included in the pivotal study is weak.

Several *in vitro* studies have been conducted to help characterize the pharmacokinetic properties of blarcamesine. Studies in human liver microsomes showed that blarcamesine is metabolized to ANAVEX19-144 by cytochrome P450 (CYP) 3A4/5 (35.3%), 2C19 (31.6%), and 2B6 (19.3%). ANAVEX19-144 is the only major metabolite of blarcamesine expected based on preclinical results.

Due to the prespecified flexible dosing design of study ANAVEX2-73-AD-004, the 30 mg and 50 mg assigned dosage arms reached, according to the applicant, similar average daily doses and corresponding similar exposures for blarcamesine and ANAVEX19-144 (table below).

Table 2. Average daily doses and corresponding exposures for Blarcamesine and ANAVEX19-144

Active Treatment Group	Statistic	Week 48	Week 48	Week 48
		Average Daily Dose	Average PK Blarcamesine (ng/mL)*	Average PK ANAVEX19-144 (ng/mL)*
Blarcamesine up to 30 mg	n	113	110	110
	Mean (SD)	**25.7** (7.29)	4.796 (3.134)	26.633 (14.24)
	Median	30	4.085	24.305
Blarcamesine up to 50 mg	n	92	87	87
	Mean (SD)	**27.8** (14.96)	5.817 (4.55)	31.031 (20.514)
	Median	30	4.372	27.28

ER analyses confirmed, according to the applicant, the ER relationship for efficacy relative to the treatment groups (placebo and combined blarcamesine) and confirms the best tolerated dose achieved based on Study ANAVEX2-73-AD-004.

A longitudinal linear efficacy model was developed to describe the relationship between drug exposure either based on plasma exposure of blarcamesine and ANAVEX19-144 or categorical (on or off drug), relative to the effects on slowing disease progression for ADAS-Cog13, CDR-SB, and ADCS-ADL over the 48-week treatment period. The longitudinal linear efficacy model using plasma exposure of blarcamesine and ANAVEX19-144 was unstable. Therefore, a categorical response was used for the final efficacy model. The efficacy model described the observed results and attempted to show that there was a reduction in disease progression with blarcamesine. Covariates including age, renal impairment, hepatic impairment, sex, and S1R/SIGMAR1 mutation status were not significant predictors of response for the efficacy model.

The most common Grade 3 or greater TEAEs were within the CNS system organ class. To further support titration with respect to safety, ER relationships for the effectiveness of titration on potentially reducing Grade 3 or higher TEAEs were explored for Study ANAVEX2-73-AD-004.

The applicant is claiming that due to the flexible dosing design of the study, the 30mg and 50mg groups reached similar average daily doses and similar exposures for blarcamesine and the main metabolite ANAVEX19-144. However, there are tolerability issues which led patients in the 50mg group to have their doses lowered or they even discontinued the study (only 66.7% in the blarcamesine 30mg group and 53.3% in the blarcamesine 50mg group completed the study vs 80% in the placebo group). Tt cannot be agreed that the exposure levels achieved a broad therapeutic window. It is also observed that patients in the so-called "blarcamesine 50mg group" had a mean average daily dose of 27.8mg, which is 55% of the maintenance dose aimed initially.

2.6.5.2. Main study(ies)

ANAVEX2-73-AD-004> A Phase 2b/3, Double-Blind, Randomised, Placebo-Controlled 48-Week Safety and Efficacy Trial of ANAVEX2-73 for the Treatment of Early Alzheimer's Disease (AD)

Methods

Study ANAVEX2-73-AD-004 was a Phase 2b/3 48-week, double-blind, randomized, placebo-controlled, parallel group design study to evaluate the effects of blarcamesine on the co-primary endpoints of

cognition and function and to assess its safety and tolerability in participants aged 60 to 85 years with early AD (i.e., MCI or early-stage mild dementia due to AD). Participants were randomly assigned in a 1:1:1 ratio to 1 of 3 dose groups: blarcamesine target dose 30 mg, blarcamesine target dose 50 mg, or placebo. The titration schedule was changed during the study (see figures below).

The study included a screening period of up to 4 weeks, followed by a 48-week treatment period during which eligible participants received study drug QD. Patients who completed the 48-week treatment period were eligible to enrol in an open-label long-term extension study (ANAVEX2-73-AD-EP-004).

Figure 5. 2-Week titration schedule: study ANAVEX2-73-AD-004 initial protocol (version 1.0 and version 2.0)

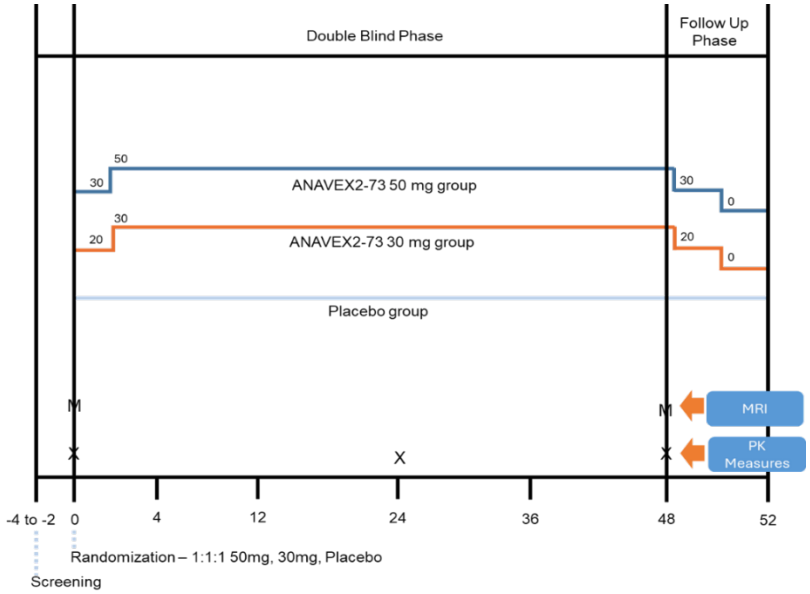
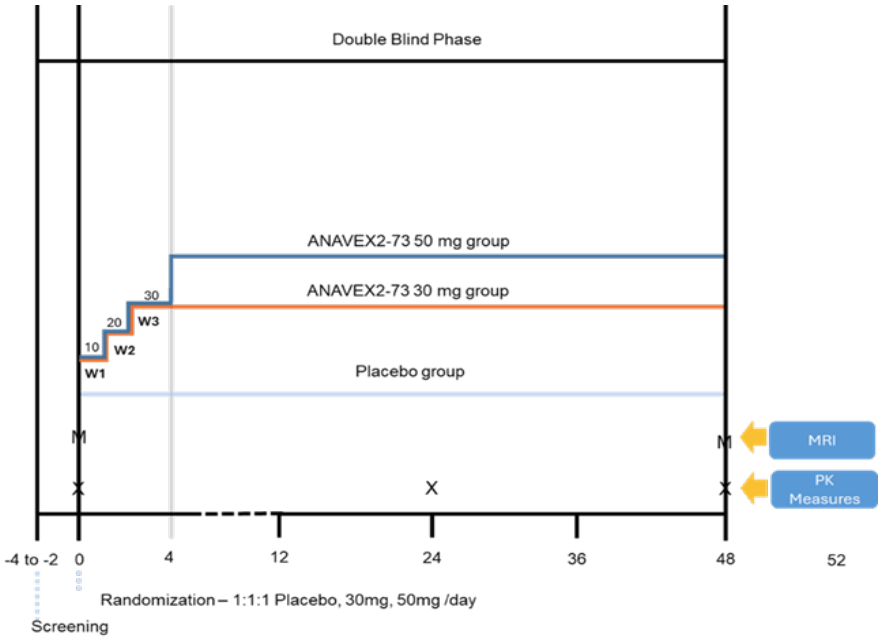


Figure 6. 3-week titration schedule: study ANAVEX2-73-AD-004 beginning with protocol version 2.1



- **Study Participants**

To be eligible for this study, participants had to meet the following key inclusion criteria:

- Be male or female aged 60 to 85 years, inclusive, with 2011 National Institute on Aging and Alzheimer's Association (NIA-AA) criteria for diagnosis of MCI due to AD or early-stage mild dementia due to AD. AD diagnosis must be made by an appropriately qualified board-certified or equivalent medical specialist.
- Have at least 1 of the following criteria to support the AD diagnosis.

- Historical records of amyloid cerebrospinal fluid (CSF) assessment, or
- Historical records of PET scan (amyloid scan or fluorodeoxyglucose [FDG]-PET), or
- Historical computed tomography (CT) or MRI scan within 18 months of screening that is consistent with a diagnosis of AD.
- MMSE score between 20-28, inclusive at both the Screening Visit and the Randomization Visit.
- Free Recall score ≤17 or Total Recall score <40 on the Free and Cued Selective Reminding Test (FCSRT).

A total of 509 participants were planned for enrolment.

The study was conducted at 55 active sites across 5 countries (Canada, 11 sites; Germany, 7 sites; United Kingdom (UK), 15 sites; The Netherlands, 3 sites; and Australia, 19 sites).

- **Treatments**

Eligible participants began treatment with study drug on Day 1 and titrated to the target dose according to the titration paradigm specified in the protocol. Originally, the dose titration was 2 weeks; the 30 mg dose group started at 20 mg/day for the first 2 weeks and then increased to 30 mg/day beginning with Week 3 and the 50 mg dose group started at 30 mg/day for the first 2 weeks and then increased to 50 mg/day beginning with Week 3. In protocol amendment 2.1, the titration period was modified to 3 weeks; both the 30 mg and 50 mg dose groups started at 10 mg/day for the first week, then increased to 20 mg/day for the second week, followed by 30 mg/day starting the third week. The 50 mg dose group then escalated to 50 mg/day starting in the fourth week. The reason for this change in dose titration schedule was the reduced tolerability observed when participants escalated dose rapidly. Beginning with the original protocol, the study also employed a flexible dosing schedule where participants were allowed to down-titrate and up-titrate based on tolerability following the initial 3-week (or 2-week) titration phase. Participants who were unable to tolerate the lowest dose (10 mg) were taken off the study drug and returned to the clinic for an Early Termination Visit. Participants who experienced any dose interruption of >10 consecutive days were withdrawn from the study. Study visits were performed every 12 weeks through Week 48.

- **Objectives**

The co-primary objectives of this study were:

- Reduction in cognitive decline assessed from baseline over 48 weeks with blarcamesine compared to placebo using the Alzheimer Disease Assessment Scale-Cognition (ADAS-Cog) scale.

  Reduction in decline of the ability to perform daily activities assessed from baseline over 48 weeks with blarcamesine compared to placebo using the Activities of Daily Living Scale (ADCS-ADL) scale.

While some of the reporting will focus on the two active doses pooled against placebo, the analyses most in line with the study design and planning (see sample size calculation below) is to compare the doses each separately against placebo, with appropriate methods to account for multiplicity.

As – during the evaluation procedure – the Applicant switched their primary focus on SIGMAR1 wild-genotype patients, results in this subgroup will be given prominence in the presentation of the trial results in the following sections.

- **Outcomes/endpoints**

According to the applicant, the efficacy assessments used in this study are standardized outcome

measures, commonly used in AD. These included scales such as:

- Alzheimer Disease Assessment Scale-Cognition (ADAS-Cog13) and
- Alzheimer Disease Cooperative Study-Activities of Daily Living Scale (ADCS-ADL)
- Clinical Dementia Rating Scale Sum of Boxes (CDR-SB)
- Mini-Mental-Status-Examination (MMSE)
- Neuropsychiatric Inventory Questionnaire (NPI-Q)
- Sleep Assessment
- Quality of Life of Alzheimer' s Disease (QoL-AD)
- Zarit Burden Interview (ZBI)
- Clinician's Global Impression – Severity and Improvement (CGI-S and CGI-I)

Structural MRI is an integral part of the clinical assessment of patients with AD.

Participants (except for participants in Australia and UK) who did not have either a CSF or PET confirmation of diagnosis of AD received a CSF assessment prior to Baseline (before the first dose of investigational product).

The safety assessments used in this study (monitoring of AEs, clinical laboratory testing, physical examinations, vital signs, and ECGs) are standardly used in clinical trials to evaluate safety of the study treatment.

- **Sample size**

Sample size and power calculations were based on a simulation approach with planned scenarios of co-primary efficacy endpoints (ADAS-Cog13 and ADCS-ADL). Bonferroni adjustment was used (for the sample size estimation) to control for the two active treatment arms versus placebo, given an overall Type I error rate of 0.05.

A sample size of 114 participants per arm was sufficient to detect a mean difference between either participants arm (i.e., 30 mg or 50 mg) and placebo of 1.5 points (SD=4.5) in the ADASCog13 with at least 90% power using a two-sample t-test with alpha = 0.05 (2-sided). Assuming the same effectiveness for ADCS-ADL, a sample size of 114 patients per treatment arm would also have a 90% power to detect a mean difference between either blarcamesine arm and placebo. Given a 90% power for each of the two co-primary efficacy endpoints, the overall power of the trial design would be at least 80%.

Assuming a 33% dropout rate (Grill 2010) due to the disease, 509 participants were planned to be enrolled into the study with the anticipation to have 342 completers in total or 114 completers per treatment arm.

- **Randomisation and Blinding (masking)**

This was a double-blinded study. Study drug and placebo were manufactured as similar capsules and were packaged similarly. The Sponsor and the study team remained blinded throughout the conduct of the study. The randomization list was retained at a restricted place until the end of the study (ie, until all data were entered into a database, the pre-analysis review of data took place, and the database was locked).

The Principal Investigator (PI) had the ability to unblind participants electronically through the Interactive Voice/Web Response System (IXRS) in the event of a medical emergency. PI notified the

Sponsor in the event that unplanned or unintentional unblinding occurred.

The Data Safety Monitoring Board (DSMB) reviewed unblinded safety data periodically throughout the study to make recommendations about study conduct if needed.

- **Statistical methods**

In this section, both the analyses as described in the latest versions of the protocol (May 2022) and in the Statistical Analysis Plan version 1.0 (November 2022) are summarised. For context, the last-patient-last-visit date is 28 June 2022. The analyses reported in the clinical study report (see below) differ from both the SAP and the protocol.

Analyses as described in the Protocol

Data will be tabulated by treatment group by study visit, with data listings provided for all data captured in the eCRF including laboratory data. On-treatment data will be assessed descriptively as both observed values and as changes from baseline. When tabulated, data will be presented using descriptive statistics. Continuous data will be summarized with the following descriptive statistics, unless otherwise noted. The descriptive statistics will comprise: number of observations, mean, standard deviation, median, minimum, and maximum; interquartile ranges. Categorical data will be summarized with frequencies and percentages.

Unless stated otherwise, statistical tests conducted will be two-sided at an α level of 0.05. Where the one- or two-sided test will be used will be specified in the SAP. When multiple comparisons are necessary, Bonferroni adjustment will be used.

As deemed appropriate, two-sample t-test will be used to test the difference between treatments if the target metric is normally distributed (symmetrical distributed at the minimum), otherwise, the nonparametric approach -Wilcoxon rank sum test will be used.

Every effort will be made to obtain required data at each scheduled evaluation from all patients who have been randomized. Missing data will not be imputed. If methods for imputation of missing data required, it will be presented in the SAP. Additionally, details of all analyses to be performed will be contained in the SAP.

The efficacy analysis will include any subject who receives at least 1 dose of study drug and at least one post-dose efficacy assessment.

Mixed effects model will be the used to analyze primary efficacy endpoints for the ANAVEX2-73 treatment effect over the time course of treatment duration. Analysis will include fixed effects of treatment, visit/study day, and baseline value as covariates. The random effect factor is subject. Two-sample t-test will be used to test the mean difference between treatment by visit if the target metric is symmetrical at a minimum. Otherwise, the nonparametric approach -Wilcoxon rank sum test will be used to test the median difference between treatment.

Descriptive statistics of the observed values and change from baseline will be presented by treatment group and visit. Continuous exploratory efficacy endpoints may also be analyzed using analysis of covariance, t-test, Wilcoxon rank sum test or as descriptive summaries. No correction to be made for multiple comparisons for exploratory efficacy endpoints. Analysis of categorical exploratory endpoints may be performed using chi-square tests.

Additional exploratory analyses may be performed to further study the effects of the treatment, as data warranted. Subgroup analyses will be performed, if data permit. Subgroup can be defined by disease severity, genotype sequence variations (rs1800866; rs113895332/rs61143203), and/or other grouping covariates.

Analyses as described in the SAP

Efficacy data will be tabulated by treatment group and by visit. When tabulated, data will be presented using descriptive statistics for both observed values and as change from baseline. Continuous data will be summarized with the following descriptive statistics unless otherwise noted: number of observations, mean, standard deviation, median, minimum, and maximum; and 95% CI, as appropriate. Categorical data will be summarized with frequencies and percentages.

The change from baseline of efficacy measures by treatment will be plotted via scatter plot, boxplot, or line plots as appropriate. Missing data will not be imputed unless otherwise documented in this SAP. No adjustment for multiplicity will be made.

Unless otherwise stated, one-sided test will be performed in the analyses proposed for the coprimary and secondary efficacy endpoints, as well as other efficacy endpoints.

Primary and secondary endpoints will be analyzed using the generalized linear modeling approach.

For exploratory endpoints, two-sample t-test will be used to test the difference between treatments if the target metric is normally distributed or N > 30 (by central limit theory), otherwise, the nonparametric approach -Wilcoxon rank sum test will be used. Jonckheere-Terpstra trend test may be performed to assess the trend in the dose-response with the change from baseline as the response variable and treatment as an ordinal classification variable.

The ordering of achieved ANAVEX2-73 dose groups will be placebo, BTD low, BTD medium and BTD high, respectively.

All efficacy analyses will be performed on the ITT population. Individual subject efficacy data listings will be presented.

Subgroup analyses based on rs1800866 genotype will be performed on all efficacy endpoints.

The co-primary efficacy endpoints are ADAS-Cog13 and ADCS-ADL. Descriptive statistics will be provided for the observed and change from baseline by randomized treatment, by BTD group and by visit for ITT and rs1800866 Gene Negative populations.

The primary analyses will be performed using the generalized linear modeling approach for ANAVEX2-73 treatment effect compared to placebo at week 48 for each of the co-primary endpoints (ADAS-Cog 13 and ADCS-ADL). Dependent variables will be ADAS-Cog 13 and ADCS-ADL treated as binary outcome variables. For ADAS-Cog13 the binary outcome dependent variable will be improved or stable cognition (patients who improve or maintain cognitive function at the end of treatment with ADAS-Cog 13 change from baseline $\leq$ 0 points) and not improved (ADAS-Cog 13 change from baseline > 0 points). Similarly, for ADCS-ADL, the binary outcome variable will be improved or stable function (patients who improve or maintain activities of daily living function at the end of treatment measured by ADCS-ADL change from baseline $\geq$ 0 points and not improved (ADCS-ADL change from baseline < 0 points). The independent variables will be treatment and baseline outcome measure (ADAS-Cog 13; ADCS-ADL). The independent treatment variable will combine all randomized treatment groups (all exposed to ANAVEX2-73), because of the dose-titration design of assigned treatment and the allowance of patients to down-titrate to a lower dose due to tolerability, and placebo. Corresponding 95% confidence intervals (CIs) and standard errors (SEs) with odds ratios from the models will be presented for each of the co-primary endpoints.

Individual subject data listings for ADAS-Cog and ADCS-ADL score will be presented.

The secondary efficacy endpoint is CDR-SB total score. Descriptive statistics will be provided for the observed and change from baseline by randomized treatment, by BTD group and by visit for ITT and rs1800866 Gene Negative populations. The analysis of CDR-SB will be performed using generalized

linear modeling for ANAVEX2-73 treatment effect compared to placebo by the end of treatment, week 48. The dependent variable will be a binary outcome variable generated from CDR-SB at the end of treatment. The binary outcome dependent variable will be improved or stable cognition/function (patients who improve or maintain cognition/function at the end of treatment measured by CDR-SB (change from baseline ≤ 0), and not improved cognition/function (CDR-SB change from baseline > 0). The independent variables will be treatment and baseline CDR-SB. The independent treatment variable will combine all randomized treatment groups (all exposed to ANAVEX2-73), because of the dose titration design of assigned treatment and the allowance of patients to down-titrate to a lower dose due to tolerability, and placebo. Corresponding 95% CIs and SEs with odds ratios from the model will be presented.

Results

- **Participant flow**

A total of 508 patients was included in the FAS. The table below shows the noteworthy discontinuation rates, especially in the active arm. The definitions of the ITT populations are found in the section below the numbers analysed.

Table 3. Study ANAVEX2-73-AD-004, participants' flow by treatment arm

	Placebo n (%)	Blarcamesine 30 mg/day n (%)	Blarcamesine 50 mg/day n (%)	Active Total n (%)
Participants in FAS Population[a]	170	169	169	338
Participants in Safety Population	168 (98.8)	167 (98.8)	168 (99.4)	335 (99.1)
Participants in ITT Population	164 (96.5)	154 (91.1)	144 (85.2)	298 (88.2)
Participants in RS-Negative Gene Population	101 (59.4)	106 (62.7)	93 (55.0)	199 (58.9)
Participants in RS- Homozygous Negative Population	152 (89.4)	149 (88.2)	130 (76.9)	279 (82.5)
Participants in PK population	NA	160 (94.7)	162 (95.9)	322 (95.3)
Completed Study	**136 (80.0)**	**112 (66.3)**	**90 (53.3)**	**202 (59.8)**
Discontinued Study Early	34 (20.0)	57 (33.7)	79 (46.7)	136 (40.2)
Discontinued Prior to Study Treatment	2 (1.2)	2 (1.2)	1 (0.6)	3 (0.9)
Reason for Early Study Discontinuation[b]				
Adverse event	13 (38.2)	41 (71.9)	65 (82.3)	106 (77.9)
Withdrawal of consent	7 (20.6)	12 (21.1)	7 (8.9)	19 (14.0)
Protocol violation	6 (17.6)	0	2 (2.5)	2 (1.5)
Death[c]	0	0	1 (1.3)	1 (0.7)
Lost to follow up	2 (5.9)	1 (1.8)	1 (1.3)	2 (1.5)
Other	6 (17.6)	3 (5.3)	3 (3.8)	6 (4.4)
Discontinued Study Early in Titration Phase	7 (4.1)	19 (11.2)	15 (8.9)	34 (10.1)
Reasons for Discontinuation from Study during Titration Phase[d]				
Adverse Event	3 (42.9)	15 (78.9)	14 (93.3)	29 (85.3)
Withdrawal of consent	1 (14.3)	4 (21.1)	0	4 (11.8)
Protocol Violation	2 (28.6)	0	0	0
Death	0	0	0	0
Lost to follow up	0	0	1 (6.7)	1 (2.9)
Other	1 (14.3)	0	0	0
Discontinued Study Early in Maintenance Phase	25 (14.7)	36 (21.3)	63 (37.3)	99 (29.3)
Reasons for Discontinuation from Study during Maintenance Phase[e]				
Adverse Event	10 (40.0)	26 (72.2)	51 (81.0)	77 (77.8)
Withdrawal of consent	6 (24.0)	7 (19.4)	7 (11.1)	14 (14.1)
Protocol Violation	4 (16.0)	0	2 (3.2)	2 (2.0)
Death	0	0	1 (1.6)	1 (1.0)
Lost to follow up	2 (8.0)	1 (2.8)	0	1 (1.0)
Other	3 (12.0)	2 (5.6)	2 (3.2)	4 (4.0)

Abbreviations: FAS = Full Analysis Set; ITT = Intent-to treat; PK = Pharmacokinetic
Percentages based on participants in FAS Population.
[a] Five participants did not have enough data to fulfill the criterion of 'enrolled participants' but were randomized and are included under FAS population. Three participants (1 in each treatment group) were randomized in error; 1 participant in the placebo group was withdrawn at the discretion of the investigator; 1 participant in the blarcamesine 30 mg group withdrew consent
[b] Percentages are based upon those who discontinued early rather than the FAS population.
[c] One participant in the placebo group died during the OLE study. The participant originated treatment with placebo and rolled over into the OLE, initiating treatment with blarcamesine. The participant died 115 days after completing the DB study and the death was assessed as unrelated to treatment. This death is not recorded in this disposition table as a result. The death is discussed in Section 12.3.2 because the TEAE that led to death began during the DB study.
[d] Percentages are based upon those who discontinued early during the titration phase.
[e] Percentages are based upon those who discontinued early during the maintenance phase.

- **Recruitment**

Start Date: 27 Aug 2018 (FPFV)

Stop Date: 28 Jun 2022 (LPLV)

- **Conduct of the study**

The applicant made several changes to the definition of the analysis set (see above) and to the analysis. The primary analysis – according to the SAP – was supposed to be based on dichotomised outcomes using a generalized linear modeling approach. This was changed to a mixed model for repeated measures (MMRM) for the continuous endpoints.

Also compared to the analysis described in the study protocol (the last versions thereof issued before SAP), the analysis was changed. The latest version of the protocol specified a set of covariates different from the set used in the analysis presented in the CSR, and did not specify the covariance structure to be used. Whereas the standard choice would be the unstructured covariance matrix (that was also specified in previous versions of the protocol), the applicant selected a Toeplitz covariance structure, which, as discussed below, lowers the p-value in this case.

- **Baseline data**

Demographic data for the FAS population are reported in the table below. Gender was similar across assigned dose groups with a somewhat higher proportion of men than women. The presence of *SIGMAR1 rs1800866* gene variant was distributed similarly across dose groups. BMI was similar across dose groups. Demographics for the ITT population were similar to the FAS population.

Table 4. Study ANAVEX2-73-AD-004, demographic and baseline characteristics by treatment group (FAS population)

Characteristic	Placebo N=170	Blarcamesine (30 mg) N=169	Blarcamesine (50 mg) N=169	Active Total N=338
Age (years)				
n	170	169	169	338
Mean (SD)	73.4 (6.44)	73.9 (6.76)	73.9 (6.49)	73.9 (6.62)
Median (min, max)	73.0 (56, 86)	74.0 (56, 86)	74.0 (58, 87)	74.0 (56, 87)
Gender, n (%)				
Female	83 (48.8)	83 (49.1)	77 (45.6)	160 (47.3)
Male	87 (51.2)	86 (50.9)	92 (54.4)	178 (52.7)
Ethnicity, n (%)				
Hispanic/Latino/Spanish origin	2 (1.2)	6 (3.6)	2 (1.2)	8 (2.4)
Not Hispanic/Latino/Spanish origin	158 (92.9)	156 (92.3)	160 (94.7)	316 (93.5)
Not disclosed	10 (5.9)	7 (4.1)	7 (4.1)	14 (4.1)
Race, n (%)				
White	163 (95.9)	165 (97.6)	164 (97.0)	329 (97.3)
Asian	2 (1.2)	3 (1.8)	5 (3.0)	8 (2.4)
Black/African American	2 (1.2)	0	0	0
American Indian or Alaska Native	0	0	0	0
Native Hawaiian or Other Pacific Islander	0	0	0	0
Other	3 (1.8)	1 (0.6)	0	1 (0.3)
***rs1800866* Gene Variant, n (%)**				
Absence	106 (62.4)	117 (69.2)	110 (65.1)	227 (67.2)
Presence	59 (34.7)	49 (29.0)	48 (28.4)	97 (28.7)
Missing	5	3	11	14
BMI (kg/m^2)				
n	165	166	167	333
Mean (SD)	25.61 (3.851)	25.57 (3.454)	25.43 (4.132)	25.50 (3.804)
Median (min, max)	25.10 (18.5, 37.2)	25.20 (17.4, 35.2)	25.20 (18.3, 41.8)	25.20 (17.4, 41.8)

Abbreviations: BMI= Body Mass Index, BTD=Best Tolerated Dose; FAS=Full Analysis Set; Max=Maximum; Min=Minimum; SD=Standard Deviation
N=the total number of participants in each group
n=the number of participants with non-missing data for the specific characteristic
Percentages are based on N.
BMI is calculated by BMI = weight (kg)/ [height (cm)/100]2
Presence of *SIGMAR1 rs1800866* Gene Variant is defined as all participants for whom *SIGMAR1 rs1800866* genotyping results in Heterozygous T/G (Heterozygous Mutant) or Homozygous G/G (Homozygous Variant [Mutant]) whereas Absence of *SIGMAR1 rs1800866* Gene Variant is defined as all participants for whom *rs1800866* genotyping results in Homozygous T/T (wild type).

Baseline disease characteristics are provided in the following tables, first for the primary ITT population and subsequently for the two genetically-defined subgroups. Baseline disease characteristics of participants were compatible with an early AD population as evidenced by MMSE >20 and elevated baseline levels of plasma p-Tau (181) and p-Tau (231).

Table 5. Study ANAVEX2-73-AD-004 clinical study report, disease characteristics by treatment group (ITT population)

Characteristic	Placebo N=164	Blarcamesine (30 mg) N=154	Blarcamesine (50 mg) N=144	Active Total N=298
MMSE				
n	164	154	144	298
Mean (SD)	23.0 (2.65)	23.6 (3.08)	23.6 (2.81)	23.6 (2.95)
Median (min, max)	23.0 (17, 30)	24.0 (14, 29)	24.0 (15, 29)	24.0 (14, 29)
Abeta42/Abeta40 ratio				
n	101	103	97	200
Mean (SD)	0.1464 (0.05193)	0.1501 (0.07081)	0.1824 (0.14854)	0.1658 (0.11608)
Median (min, max)	0.1363 (0.081, 0.395)	0.1377 (0.084, 0.755)	0.1431 (0.104, 1.143)	0.1379 (0.084, 1.143)
NfL (ng/L)				
n	159	152	140	292
Mean (SD)	21.86 (8.294)	21.95 (11.174)	21.26 (9.065)	21.62 (10.206)
Median (min, max)	20.34 (6.9, 53.3)	19.79 (7.4, 80.6)	19.23 (8.5, 64.1)	19.57 (7.4, 80.6)
Plasma p-Tau (231)				
n	123	102	97	199
Mean (SD), ng/L	27.08 (34.579)	29.02 (29.546)	34.19 (50.757)	31.54 (41.244)
Median (min, max)	17.93 (2.0, 287.8)	20.55 (3.8, 213.7)	19.98 (6.1, 472.6)	20.38 (3.8, 472.6)
Plasma p-Tau (181)				
n	153	145	132	227
Mean (SD), ng/L	65.42 (28.046)	61.88 (25.438)	62.62 (25.752)	62.23 (25.545)
Median (min, max)	62.57 (16.6, 160.8)	61.59 (17.3, 141.8)	57.95 (19.9, 152.8)	59.95 (17.3, 152.8)
ADAS-Cog13				
n	164	154	144	298
Mean (SD)	30.4 (8.44)	28.4 (8.44)	28.9 (9.08)	28.6 (8.74)
Median (min, max)	30.0 (13, 69)	29.0 (7, 56)	28.5 (10, 66)	29.0 (7, 66)
ADCS-ADL				
n	164	154	144	298
Mean (SD)	66.4 (7.09)	66.7 (7.36)	67.0 (7.94)	66.9 (7.63)
Median (min, max)	67.0 (37, 78)	68.0 (42, 78)	69.0 (30, 78)	69.0 (30, 78)
CDR-SB Total Score				
n	164	154	144	298
Mean (SD)	4.1 (1.76)	3.8 (1.63)	3.8 (1.81)	3.8 (1.72)
Median (min, max)	3.8 (1, 9)	3.5 (1, 10)	3.5 (1, 10)	3.5 (1, 10)
AD Prior Medication (Safety Population)				
N	168	167	168	335
Yes, n (%)	111 (66.1)	109 (65.3)	109 (64.9)	218 (65.1)

Abbreviations: AD = Alzheimer's disease; ITT = Intent to Treat; Max=Maximum; Min=Minimum; MMSE = Mini Mental State Examination; NfL=neurofilament light chain; SD=Standard Deviation
N=the total number of participants in each group
n=the number of participants with non-missing data for the specific characteristic Percentages are based on N.

Table 6. Demographics for the AD-004 gene variant subgroup populations ITT population

	SIGMAR1/rs1800866 WT +/+ Population				**SIGMAR1(rs1800866)/COL24A1(rs60891279) WT +/+ Population**			
Demographic characteristics	Blarcamesine 30 mg (N=106)	Blarcamesine 50 mg (N=93)	Active Total (N= 199)	Placebo (N= 101)	Blarcamesine 30 mg (N=79)	Blarcamesine 50 mg (N=77)	Active Total (N=156)	Placebo (N=66)
Sex, n (%)								
Female	53 (50.0)	45 (48.4)	98 (49.2)	52 (51.5)	40 (50.6)	41 (53.2)	81 (51.9)	33 (50.0)
Male	53 (50.0)	48 (51.6)	101 (50.8)	49 (48.5)	39 (49.4)	36 (46.8)	75 (48.1)	33 (50.0)
Age, Mean (SD)	73.3 (6.94)	74.0 (6.05)	73.6 (6.54)	72.9 (6.21)	73.7 (7.24)	73.6 (5.86)	73.7 (6.57)	72.9 (6.18)
Race, n (%)								
Asian	1 (0.9)	2 (2.2)	3 (1.5)	1 (1.0)	0	2 (2.6)	2 (1.3)	0
ack or African American	0	0	0	1 (1.0)	0	0	0	1 (1.5)
Other	0	0	0	2 (2.0)	0	0	0	1 (1.5)
White	105 (99.1)	91 (97.8)	196 (98.5)	97 (96.0)	79 (100.0)	75 (97.4)	154 (98.7)	64 (97.0)
Ethnicity, n (%)								
ispanic or Latino/a or of Spanish origin	3 (2.8)	0	3 (1.5)	0	3 (3.8)	0	3 (1.9)	0
Not Disclosed	5 (4.7)	6 (6.5)	11 (5.5)	6 (5.9)	4 (5.1)	5 (6.5)	9 (5.8)	3 (4.5)
ot Hispanic or Latino/a or of Spanish origin	98 (92.5)	87 (93.5)	185 (93.0)	95 (94.1)	72 (91.1)	72 (93.5)	144 (92.3)	63 (95.5)
APOE e4 genotype, n (%)								
E2/E3	5 (4.7)	4 (4.3)	9 (4.5)	4 (4.0)	5 (6.3)	3 (3.9)	8 (5.1)	4 (6.1)
E2/E4	2 (1.9)	2 (2.2)	4 (2.0)	5 (5.0)	1 (1.3)	2 (2.6)	3 (1.9)	2 (3.0)
E3/E3	25 (23.6)	28 (30.1)	53 (26.6)	20 (19.8)	19 (24.1)	23 (29.9)	42 (26.9)	15 (22.7)
E3/E4	45 (42.5)	39 (41.9)	84 (42.2)	47 (46.5)	33 (41.8)	33 (42.9)	66 (42.3)	28 (42.4)
E4/E4	23 (21.7)	16 (17.2)	39 (19.6)	21 (20.8)	17 (21.5)	12 (15.6)	29 (18.6)	15 (22.7)
Missing	6 (5.7)	4 (4.3)	10 (5.0)	4 (4.0)	4 (5.1)	4 (5.2)	8 (5.1)	2 (3.0)
Baseline clinical scores, Mean (SD)								
ADAS-COG 13 score	28.6 (8.51)	29.3 (9.68)	28.9 (9.06)	29.9 (8.87)	28.4 (9.10)	29.3 (10.00)	28.8 (9.54)	30.4 (9.23)
ADCS-ADL score	65.9 (7.53)	67.9 (7.42)	66.8 (7.53)	67.1 (6.47)	65.3 (7.68)	67.8 (7.71)	66.5 (7.77)	66.8 (6.01)
CDR-SB score	3.96 (1.708)	3.66 (1.819)	3.82 (1.763)	3.95 (1.727)	4.02 (1.853)	3.66 (1.903)	3.84 (1.880)	4.02 (1.699)
MMSE score	23.6 (3.17)	23.6 (2.84)	23.6 (3.02)	23.2 (2.79)	23.5 (3.21)	23.4 (2.96)	23.5 (3.08)	23.0 (2.55)
Baseline CDR-Global scores, n (%)								
0.5	61 (57.5)	63 (67.7)	124 (62.3)	61 (60.4)	43 (54.4)	51 (66.2)	94 (60.3)	39 (59.1)
1	43 (40.6)	29 (31.2)	72 (36.2)	39 (38.6)	34 (43.0)	25 (32.5)	59 (37.8)	26 (39.4)
2	1 (0.9)	1 (1.1)	2 (1.0)	1 (1.0)	1 (1.3)	1 (1.3)	2 (1.3)	1 (1.5)
3	1 (0.9)	0	1 (0.5)	0	1 (1.3)	0	1 (0.6)	0

	SIGMAR1/rs1800866 WT +/+ Population				SIGMAR1(rs1800866)/COL24A1(rs60891279) WT +/+ Population			
Demographic characteristics	Blarcamesine 30 mg (N=106)	Blarcamesine 50 mg (N=93)	Active Total (N= 199)	Placebo (N= 101)	Blarcamesine 30 mg (N=79)	Blarcamesine 50 mg (N=77)	Active Total (N=156)	Placebo (N=66)
MMSE score at baseline, n (%)								
<=20	16 (15.1)	15 (16.1)	31 (15.6)	17 (16.8)	15 (19.0)	14 (18.2)	29 (18.6)	11 (16.7)
>20	90 (84.9)	78 (83.9)	168 (84.4)	84 (83.2)	64 (81.0)	63 (81.8)	127 (81.4)	55 (83.3)
Concomitant AD medication	69 (65.1)	65 (69.9)	134 (67.3)	71 (70.3)	50 (63.3)	56 (72.7)	106 (67.9)	50 (75.8)
cetyl cholinesterase inhibitors	65 (61.3)	60 (64.5)	125 (62.8)	68 (67.3)	46 (58.2)	51 (66.2)	97 (62.2)	48 (72.7)
Memantine	13 (12.3)	7 (7.5)	20 (10.1)	10 (9.9)	8 (10.1)	6 (7.8)	14 (9.0)	7 (10.6)
Baseline Plasma p-Tau (181)								
o. of participants evaluated at baseline	99	84	183	96	74	70	144	61
aseline mean (SD), pg/mL	64.0 (26.70)	63.7 (25.77)	63.9 (26.21)	66.3 (28.82)	62.7 (28.09)	62.1 (23.82)	62.4 (26.01)	69.2 (30.25)
Baseline Plasma p-Tau (231)								
o. of participants evaluated at baseline	69	63	132	71	49	51	100	48
aseline mean (SD), pg/mL	31.6 (33.56)	37.4 (60.33)	34.4 (48.12)	24.5 (16.59)	31.6 (35.24)	39.9 (66.56)	35.8 (53.45)	23.3 (16.10)

- **Numbers analysed**

The analysis on the primary population included patients in the ITT population. In the clinical study report, this population was defined as including all randomized participants (FAS) who received at least one dose of study drug and who had baseline and at least one post-baseline efficacy assessments. It is noted that in previous versions of the Protocol the ITT population was defined as all randomised patients (which is more in line with the ITT principle).

Under the CSR definition, 164 patients treated with placebo, 154 patients treated with Blarcamesine 30 mg/day, and 144 patients treated with Blarcamesine 50 mg/day were included in the ITT population. It should be noted that the difference in the number of subjects who received at least one dose of study drug and ITT are for the placebo n=4 and for the blarcamesine active total n=37 (for the blarcamesine 30 mg group n=13 and for the 50mg group n=24).

During the application procedure, the applicant presented analyses focusing on the subgroups of patients who are homozygous wild-type for SIGMAR1 (rs1800866) and for both SIGMAR1 and COL24A1 (rs60891279).

The study AD-004 gene variant subgroup populations were as follows:

Table 7. Numbers of patients included in the main subgroups investigated

SIGMAR1/rs1800866 WT +/+ Population				SIGMAR1(rs1800866)/COL24A1(rs60891279) WT +/+ Population			
Blarcamesine 30 mg (N=106)	Blarcamesine 50 mg (N=93)	Active Total (N=199)	Placebo (N=101)	Blarcamesine 30 mg (N=79)	Blarcamesine 50 mg (N=77)	Active Total (N=156)	Placebo (N=66)

- **Outcomes and estimation**

Co-primary endpoints: ADAS-Cog13 and ADCS-ADL

According to the applicant, for one of the two co-primary endpoints, ADAS-Cog13, treatment with blarcamesine (prespecified combined dose groups) for 48 weeks resulted in a significantly lower least squares (LS) mean change from baseline at Week 48 (3.555) compared to placebo (5.582), resulting in a LS mean difference of -2.027 (P = 0.0079) for active vs. placebo in favour of the active treatment, and individual active groups also demonstrated significantly slowed decline relative to placebo at 48 weeks: 30 mg group difference in LS mean change from baseline of -1.934 (P = 0.0263); 50 mg group difference in LS mean change from baseline of -2.149 (P = 0.0214).

Table 8. MMRM analysis: change from baseline in co-primary efficacy endpoint ADAS-Cog13 (ITT population) – study ANAVEX2-73-AD-004

	Individual Group Comparison			Group Comparison	
Visit Parameter	**Placebo (N = 164)**	**30 mg (N = 154)**	**50 mg (N = 144)**	**Placebo (N = 164)**	**Active Total (N = 298)**
Week 48					
N	122	108	83	122	191
LS Mean (SE)	5.584 (0.855)	3.650 (0.883)	3.436 (0.976)	5.582 (0.853)	3.555 (0.792)
LS Mean Diff (SE)	N/A	-1.934 (0.869)	-2.149 (0.932)	N/A	-2.027 (0.761)
95% CI of LS Mean Diff	N/A	(-3.639, -0.228)	(-3.979, -0.319)	N/A	(-3.522, -0.533)
p-value	N/A	0.0263	0.0214	N/A	0.0079

Abbreviations: CI=confidence interval; diff=difference; LS=least squares; MMRM=Mixed Model Repeated Measures; N/A=not applicable; SE=Standard Error

Due to concerns in the definition of the analysis population (see above) and the handling of missing data (see below), the Applicant was asked to produce an analysis with a more standard definition of ITT population ("Randomisation population" in the submission) and with control-based missing data imputation. The results are below and – also for ADAS-Cog13 – they are no longer statistically significant.

Table 9. Analysis of ADAS-Cog13 at week 48 - MMRM model without RSGENEN and ln(Nf-L) covariates randomization population (pattern mixture model with control-based pattern imputation)

	Individual Group Comparison				Combined Group Comparison
	Placebo (N=170)	Blarcamesine (30 mg) (N=169)	Blarcamesine (50 mg) (N=169)	Placebo (N=170)	Active Total (N=338)
Change from Baseline at Week 48 MMRM [1]					
n	168	167	168	168	335
LS Mean (SE)	5.075 (0.842)	3.660 (0.867)	3.750 (0.920)	5.072 (0.841)	3.703 (0.770)
95% CI of LS Mean	(3.424, 6.726)	(1.959, 5.361)	(1.944, 5.556)	(3.422, 6.722)	(2.192, 5.214)
LS Mean Diff (SE)	N/A	-1.414 (0.850)	-1.325 (0.898)	N/A	-1.369 (0.749)
95% CI of LS Mean Diff	N/A	(-3.082, 0.253)	(-3.087, 0.438)	N/A	(-2.837, 0.099)
p-value	N/A	0.0964	0.1406	N/A	0.0676

LS = least squares; SE = standard error; N/A = not applicable .
N = The total number of subjects in each group under the specific population. n = Number of subjects with non-missing data for the specific Category/Parameter and model covariates.
[1] Mixed Models for Repeated Measures (MMRM) model is used and include treatment group (one model include all treatments in individual group comparison, the other include treatments for combined group comparison), visit, country, and treatment group by visit interaction as fixed effects, the baseline of ADAS-Cog13, whether receiving concomitant AD medication at baseline, baseline MMSE total score (>20 cutoff) as covariate based on Toeplitz covariance structure matrix and Kenward-Roger corrected degree of freedom.

The Applicant also performed tipping point analysis. Given the amount of patients who discontinue blarcamesine, of particular interest are the set of tipping point analyses in which participants on blarcamesine who have reached at least the first scheduled visit had imputed values over a range of possible negative treatment effects. For active group participants, missing values would need to shift 1.9 points in ADAS-Cog13 score to reach the tipping point where the result is no longer significant.

For the other co-primary endpoint, ADCS-ADL, no statistically significant difference was shown in the primary analysis presented.

Table 10. Change from baseline in co-primary efficacy endpoint ADCS-ADL by visit (ITT population)

	Individual Group Comparison			Group Comparison	
	Placebo (N = 164)	**30 mg (N = 154)**	**50 mg (N = 144)**	**Placebo (N = 164)**	**Active Total (N = 298)**
Week 48 (Co-Primary Endpoint)					
n	126	109	85	126	194
LS Mean (SE)	-7.592 (0.928)	-6.702 (0.971)	-6.940 (1.071)	-7.560 (0.928)	-6.785 (0.869)
LS Mean Diff (SE)	N/A	**0.890** (0.959)	**0.652** (1.030)	N/A	**0.775** (0.840)
95%CI of LS Mean Diff	N/A	(-0.992, 2.772)	(-1.370, 2.673)	N/A	(-0.874, 2.423)
p-value	N/A	**0.3535**	**0.5273**	N/A	**0.3565**

Abbreviations: CI = confidence interval; ITT = intent-to-treat; LS = least squares; MMRM = mixed model for repeated measures; SE = Standard Error; * p < 0.05; ** p < 0.01

Secondary endpoint: CDR-SB

According to the applicant, the results on ADAS-Cog13 are supported by findings on the CDR-SB, a global measure of cognition and function and secondary endpoint. CDR-SB had significantly less decline in the pooled blarcamesine group than in the placebo group (LS mean difference of -0.483, P = 0.010). Significant difference from placebo was also observed for both 50 mg (LS mean difference of -0.465, P = 0.045) and 30 mg (LS mean difference of -0.502, P = 0.020) dose groups.

Table 11. Change from baseline in secondary efficacy endpoint CDR-SB (ITT population) – study ANAVEX2-73-AD-004

	Individual Group Comparison			Group Comparison	
Visit Parameter	**Placebo (N = 164)**	**30 mg (N = 154)**	**50 mg (N = 144)**	**Placebo (N = 164)**	**Active Total (N = 298)**
Week 48					
n	126	107	84	126	191
LS Mean (SE)	1.755 (0.207)	1.253 (0.217)	1.290 (0.239)	1.749 (0.207)	1.266 (0.193)
LS Mean Diff (SE)	N/A	**-0.502** (0.215)	-0.465 (0.231)	N/A	**-0.483** (0.188)
95% CI of LS Mean Diff	N/A	(-0.924, -0.080)	(-0.918, -0.012)	N/A	(-0.853, -0.114)
p-value	N/A	**0.0199**	0.0445	N/A	**0.0104**

Abbreviations: CFB=Change from baseline; CI=confidence interval; diff=difference; LS=least squares; MMRM=Mixed Model Repeated Measures; N/A=not applicable; SD=standard deviation; SE=Standard

According to the applicant, a statistically significant less decline in global disease severity (CGI-I) by investigator's judgment at 48 weeks relative to baseline disease severity was observed for participants who received blarcamesine compared to those who received placebo (CGI-I LS mean difference of -0.278 [95% CI -0.466 to -0.089]; P = 0.004), as well as both 50 mg (CGI-I LS mean difference of -0.314 [95%CI -0.545 to -0.082]; P = 0.008) and 30 mg (difference of -0.248 [95%CI -0.464 to -0.033]; P = 0.024) groups (see table below).

Table 12. CGI-I efficacy endpoint (ITT population) – study ANAVEX2-73-AD-004

Visit Parameter	Individual Group Comparison			Group Comparison	
	Placebo (N = 164)	30 mg (N = 154)	50 mg (N = 144)	Placebo (N = 164)	Active Total (N = 298)
Week 48					
n	125	107	83	125	190
LS Mean (SE)	4.882 (0.099)	4.634 (0.105)	4.568 (0.116)	4.883 (0.099)	4.606 (0.091)
LS Mean Diff (SE)	N/A	**-0.248** (0.110)	-0.314 (0.118)	N/A	-0.278 (0.096)
95% CI of LS Mean Diff	N/A	(-0.464, -0.033)	(-0.545, -0.082)	N/A	(-0.466, -0.089)
p-value	N/A	**0.0239**	0.0079	N/A	0.0039

Abbreviations: CI=confidence interval; diff=difference; LS=least squares; MMRM=Mixed Model Repeated Measures; N/A=not applicable; SE=Standard Error

Exploratory QoL and caregiver's burden

Another exploratory efficacy endpoint was to evaluate the effect of blarcamesine on participants and caregiver Quality of Life measured by the QoL-AD compared to placebo over 48-week treatment duration. The results at week 48 are shown in the table below.

Table 13. Analysis of change from baseline in QoL-AD by visit, MMRM Model, ITT population, Week 48

Visit Parameter	Individual Group Comparison			Group Comparison	
	Placebo (N = 164)	30 mg (N = 154)	50 mg (N = 144)	Placebo (N = 164)	Active Total (N = 298)
Week 48					
n	126	110	84	126	194
LS Mean (SE)	-3.289 (0.565)	-3.202 (0.586)	-2.230 (0.649)	-3.310 (0.565)	-2.808 (0.527)
LS Mean Diff (SE)	N/A	0.087 (0.577)	1.060 (0.621)	N/A	0.502 (0.506)
95% CI of LS Mean Diff	N/A	(-1.045, 1.219)	(-0.159, 2.279)	N/A	(-0.492, 1.495)
p-value	N/A	**0.8803**	0.0883	N/A	0.3221

Abbreviations: CI=confidence interval; diff=difference; LS=least squares; MMRM=Mixed Model Repeated Measures; N/A=not applicable; SE=Standard Error

The caregiver's burden was also measured through the ZBI. The results at week 48 are reported below.

Table 14. Analysis of change from baseline in ZBI by visit, MMRM model, ITT population, week 48

Visit Parameter	Individual Group Comparison			Group Comparison	
	Placebo (N = 164)	30 mg (N = 154)	50 mg (N = 144)	Placebo (N = 164)	Active Total (N = 298)
Week 48					
n	126	108	84	126	192
LS Mean (SE)	3.310 (1.080)	5.141 (1.121)	3.123 (1.239)	3.379 (1.080)	4.331 (1.011)
LS Mean Diff (SE)	N/A	1.831 (1.089)	-0.187 (1.172)	N/A	0.951 (0.955)
95% CI of LS Mean Diff	N/A	(-0.306, 3.968)	(-2.488, 2.114)	N/A	(-0.924, 2.826)
p-value	N/A	**0.0931**	0.8735	N/A	0.3196

Abbreviations: CI=confidence interval; diff=difference; LS=least squares; MMRM=Mixed Model Repeated Measures; N/A=not applicable; SE=Standard Error

Exploratory biomarker results

According to the applicant, support to the positive trends observed is offered by significant changes in physiological markers of AD. The results, are shown in the tables below.

Table 15. Analysis of plasma biomarkers related to AD pathophysiology at week 48 – T-test with Welch option (ITT population) – study ANAVEX2-73-AD-004

Biomarker Visit Parameter	Individual Group Comparison			Group Comparison	
	Placebo (N = 164)	30 mg (N = 154)	50 mg (N = 144)	Placebo (N = 164)	Active Total (N = 298)
Change from Baseline at Week 48					
Aβ42/40 ratio					
n	78	67	52	78	119
Mean (SD)	-0.0003 (0.03499)	0.0063 (0.02790)	0.0210 (0.07997)	-0.0003 (0.03499)	0.0127 (0.05703)
p-value	N/A	0.2057	0.0751	N/A	0.0480
Nf-L, ng/L					
n	122	99	83	122	182
Mean (SD)	4.92 (32.999)	1.38 (6.542)	1.96 (6.877)	4.92 (32.999)	1.65 (6.685)
p-value	N/A	0.2490	0.3385	N/A	0.2813
p-Tau 181, ng/L					
n	117	92	73	117	165
Mean (SD)	10.85 (85.625)	2.92 (14.170)	5.17 (16.329)	10.85 (85.625)	3.92 (15.157)
p-value	N/A	0.3271	0.4873	N/A	0.3884
p-Tau 231, ng/L					
n	83	50	55	83	105
Mean (SD)	3.86 (40.659)	2.27 (13.539)	-1.51 (12.081)	3.86 (40.659)	0.29 (12.874)
p-value	N/A	0.7441	0.2609	N/A	0.4432

Abbreviations: N/A=not applicable; SD=standard deviation

Table 16. ANAVEX2-73-AD-004 study, structural MRI results, general linear model with active doses combined: annualized percent change from baseline to week 48 (ITT population)

Structural MRI volume, Annualized percent change	N	LS Mean (SE)	LS Mean Difference vs Placebo	2-sided 95% CI for LS Mean Difference	p-value
Whole brain volume					
Placebo	100	-2.04% (0.23)	--	--	--
30 mg & 50 mg	165	-1.27% (0.19)	0.77%	(0.29, 1.25)	0.0019**
Total brain grey matter					
Placebo	100	-2.18% (0.44)	--	--	--
30 mg & 50 mg	166	-0.80% (0.37)	1.38%	(0.46, 2.30)	0.0035**
Total brain white matter					
Placebo	100	-1.89% (0.38)	--	--	--
30 mg & 50 mg	165	-1.98% (0.33)	-0.09%	(-0.90, 0.73)	0.832
Lateral Ventricles					
Placebo	100	10.76% (0.78)	--	--	--
30 mg & 50 mg	165	8.06% (0.67)	-2.70%	(-4.36, -1.05)	0.0015**

*: $p < 0.05$; **: $p < 0.01$ 1

2.6.5.3. Subgroup analysis

As a prespecified exploratory endpoint of the double-blind pivotal study (ANAVEX2-73-AD-004), ADAS-Cog13, ADCS-ADL, CDR-SB, and CGI-I were analysed for the following subgroups:

- the absence or presence of a S1R/SIGMAR1 gene variant (rs1800866)
- gender (male or female),

- age (≤75 or >75 years), and
- country (Canada, Australia, Netherlands, Great Britain, or Germany).

Subgroup analysis based on the SIGMAR1 status

Given the mechanism of action of blarcamesine, activation of the S1R/SIGMAR1, prespecified analyses by subgroup of participants without the S1R/SIGMAR1 gene variant rs1800866, i.e., those with a wild type (WT; T/T) genotype, compared with participants who were carriers of the gene variant was particularly highlighted during the procedure. Approximately 70% of the overall population are WT, while ~30% carry the mutated S1R/SIGMAR1 gene variant rs1800866 (T>G mutation; Laurini 2016).

Results for the common non-variant subgroup have been reported in two ways by the Applicant: referring to the subgroup either as SIGMAR1 WT or as "rs1800866 Genotype: Absence". The Applicant confirmed that this is the same subgroup, and that the differing results are due to the analysis methods: the results reported as relating to the SIGMAR1 WT subgroup were performed with MMRM model, Unstructured Covariance and no co-variates, and are reported here. Results reported as relating to the as "rs1800866 Genotype: Absence" are from analysis of the same patients but with a Toeplitz covariance structure and covariate adjustment, tend to have lower p-values (generally $p<0.05$ for the analyses reported below), but are overall similar in terms of point estimates.

The results for ADAS-Cog13, ADECS-ADL, CDR-SB, and CGI-I for the SIGMAR1 WT subgroup are presented below.

Table 17. Change from baseline at week 48 - ADAS-Cog13 for the SIGMAR1 WT subpopulation

	Individual Group Comparison			**Combined Group Comparison**	
	Placebo (N=101)	Anavex2-73 (30 mg) (N=106)	Anavex2-73 (50 mg) (N=93)	Placebo (N=101)	Active Total (N=199)
BIC		5996.2			6006.2
n	83	81	58	**83**	**139**
LS Mean (SE)	4.885 (0.861)	2.926 (0.868)	3.044 (1.010)	4.884 (0.859)	2.981 (0.657)
95% CI of LS Mean	(3.188, 6.581)	(1.215, 4.637)	(1.054, 5.034)	(3.191, 6.578)	(1.685, 4.276)
LS Mean Diff (SE)	N/A	-1.959 (1.223)	-1.840 (1.327)	N/A	**-1.904** (1.082)
95% CI of LS Mean Diff	N/A	(-4.368, 0.451)	(-4.455, 0.774)	N/A	(-4.036, 0.228)
p-value	N/A	**0.1106**	**0.1668**	N/A	**0.0799**

Table 18. Change from baseline at week 48 - analysis of ADCS-ADL by visit – MMRM model for the SIGMAR1 WT subpopulation

	Individual Group Comparison			**Combined Group Comparison**	
	Placebo (N=101)	Anavex2-73 (30 mg) (N=106)	Anavex2-73 (50 mg) (N=93)	Placebo (N=101)	Active Total (N=199)
BIC		6128.8		6142.8	
n	87	81	60	**87**	**141**
LS Mean (SE)	-6.929 (0.981)	2.926 (0.868)	-7.141 (1.165)	-6.929 (0.981)	-6.457 (0.760)
95% CI of LS Mean	(-8.864, -4.994)	(-7.889, -3.918)	(-9.437, -4.844)	(-8.862, -4.996)	(-7.955, -4.959)
LS Mean Diff (SE)	N/A	1.025 (1.406)	-0.212 (1.523)	N/A	**0.472** (1.241)
95% CI of LS Mean Diff	N/A	(-1.747, 3.798)	(-3.215, 2.791)	N/A	(-1.974, 2.918)
p-value	N/A	**0.4667**	**0.8895**	N/A	**0.7038**

Table 19. Change from baseline at week 48 – CDR-SB for the SIGMAR1 WT subpopulation

	Individual Group Comparison			**Combined Group Comparison**	
	Placebo (N=101)	Anavex2-73 (30 mg) (N=106)	Anavex2-73 (50 mg) (N=93)	Placebo (N=101)	Active Total (N=199)
BIC		3453.6			3455.7
n	87	80	59	87	139
LS Mean (SE)	1.732 (0.232)	1.118 (0.239)	1.297 (0.278)	1.732 (0.232)	1.200 (0.181)
95% CI of LS Mean	(1.274, 2.190)	(0.647, 1.590)	(0.750, 1.844)	(1.275, 2.190)	(0.843, 1.557)
LS Mean Diff (SE)	N/A	**-0.614** (0.334)	-0.435 (0.362)	N/A	**-0.532** (0.295)
95% CI of LS Mean Diff	N/A	(-1.271, 0.044)	(-1.149, 0.279)	N/A	(-1.113, 0.048)
p-value	N/A	**0.0671**	0.2311	N/A	**0.0719**

Table 20. Change from baseline at week 48 - analysis of CGI-I by visit – MMRM model for the SIGMAR1 WT subpopulation

	Individual Group Comparison			**Combined Group Comparison**	
	Placebo (N=101)	Anavex2-73 (30 mg) (N=106)	Anavex2-73 (50 mg) (N=93)	Placebo (N=101)	Active Total (N=199)
BIC		2299.7		2291.6	
n	86	79	59	**86**	**138**
LS Mean (SE)	4.721 (0.096)	4.493 (0.101)	4.442 (0.116)	4.721 (0.096)	4.471 (0.076)
95% CI of LS Mean	(4.532, 4.911)	(4.295, 4.692)	(4.214, 4.670)	(4.532, 4.911)	(4.322, 4.620)
LS Mean Diff (SE)	N/A	-0.228 (0.139)	-0.279 (0.151)	N/A	**-0.250** (0.122)
95% CI of LS Mean Diff	N/A	(-0.502, 0.047)	(-0.576, 0.017)	N/A	(-0.491, -0.009)
p-value	N/A	**0.1032**	**0.0648**	N/A	**0.0419**

Subgroup analysis based on SIGMAR1 and COL24A1 status

A post-hoc analysis of the original co-primary efficacy endpoints (ADAS-Cog13 and ADCS-ADL) was conducted in the subpopulation of participants who were WT for both *SIGMAR1* and *COL24A1* to evaluate the efficacy of blarcamesine. The applicant has admitted that the analysis was post-hoc and the results are summarised below.

The MMRM analysis of ADAS-Cog 13 total score for the ITT population active total showed at 48 weeks a mean difference vs. placebo of -4.179 (95%CI: -6.512, -1.845, P=0.0005). The LS mean difference of the 30 mg dose group at 48 weeks showed a mean difference vs. placebo of -4.739 (95%CI: -7.370, -2.108, P=0.0004). For the 50mg group the LS mean difference vs placebo was -3.528 (95%CI: -6.286, -0.770, P=0.0123). Using MMRM analysis, the ADCS-ADL and CDR-SB results were nominally statistically significant in the case of active total and 30mg groups.

Table 21. ADAS-Cog13 primary efficacy - MMRM analysis, Toeplitz covariance, SIGMAR1/COL24A1 WT homozygous

	Individual Group Comparison			Combined Group Comparison	
	Placebo (N=66)	**Blarcamesine (30 mg) (N=79)**	**Blarcamesine (50 mg) (N=77)**	**Placebo (N=66)**	**Active Total (N=156)**
Change from Baseline at Week 48					
MMRM [1]					
BIC	4334.4			4350.8	
n	54	61	48	54	109
LS Mean (SE)	**5.592** (1.533)	0.853 (1.423)	2.063 (1.503)	**5.506** (1.530)	1.327 (1.289)
LS Mean Diff (SE)	N/A	-4.739 (1.338)	-3.528 (1.403)	N/A	**-4.179** (1.187)
95% CI of LS Mean Diff	N/A	(-7.370, -2.108)	(-6.286, -0.770)	N/A	(-6.512, -1.845)
p-value	N/A	0.0004	0.0123	N/A	0.0005

LS = least squares; SE = standard error; N/A = not applicable.
N = The total number of subjects in each group under the SIGMAR1/COL24A1 WT Homozygous population. n = Number of subjects with non-missing data for the specific Category/Parameter and model covariates.
[1] Mixed Models for Repeated Measures (MMRM) model is used and include treatment group (one model include all treatments in individual group comparison, the other include treatments for combined group comparison), visit, country, SIGMAR1 gene variant status, and treatment group by visit interaction as fixed effects, the baseline of ADAS-Cog13, natural log of baseline Nf-L, whether receiving concomitant AD medication at baseline, baseline MMSE total score (>20 cutoff) as covariate based on Toeplitz covariance structure matrix and Kenward-Roger corrected degree of freedom.

Table 22. ADCS-ADL primary efficacy-MMRM analysis, Toeplitz covariance, SIGMAR1/COL24A1 WT homozygous population

	Individual Group Comparison			Combined Group Comparison	
	Placebo (N=66)	**Blarcamesine (30 mg) (N=79)**	**Blarcamesine (50 mg) (N=77)**	**Placebo (N=66)**	**Active Total (N=156)**
Change from Baseline at Week 48					
MMRM [1]					
BIC	4488.6			4502.4	
n	58	60	48	58	108
LS Mean (SE)	-7.975 (1.562)	-3.730 (1.496)	-6.199 (1.552)	-8.075 (1.567)	-4.943 (1.342)
LS Mean Diff (SE)	N/A	4.245 (1.387)	1.775 (1.463)	N/A	**3.131** (1.226)
95% CI of LS Mean Diff	N/A	(1.518, 6.972)	(-1.100, 4.651)	N/A	(0.720, 5.542)
p-value	N/A	0.0024	**0.2255**	N/A	0.0111

LS = least squares; SE = standard error; N/A = not applicable.
N = The total number of subjects in each group under the SIGMAR1/COL24A1 WT Homozygous population. n = Number of subjects with non-missing data for the specific Category/Parameter and model covariates.
[1] Mixed Models for Repeated Measures (MMRM) model is used and include treatment group (one model include all treatments in individual group comparison, the other include treatments for combined group comparison), visit, country, SIGMAR1 gene variant status, and treatment group by visit interaction as fixed effects, the baseline of ADCS-ADL, natural log of baseline Nf-L, whether receiving concomitant AD medication at baseline, baseline MMSE total score (>20 cutoff) as covariate based on Toeplitz covariance structure matrix and Kenward-Roger corrected degree of freedom.

Table 23. CDR-SB primary efficacy - MMRM analysis, Toeplitz covariance (SIGMAR1/COL24A1 WT homozygous)

	Individual Group Comparison			Combined Group Comparison	
	Placebo (N=66)	**Blarcamesine (30 mg) (N=79)**	**Blarcamesine (50 mg) (N=77)**	**Placebo (N=66)**	**Active Total (N=156)**
Change from Baseline at Week 48					
MMRM [1]					
BIC	2548.4			2557.5	
n	57	60	48	57	108
LS Mean (SE)	1.880 (0.362)	0.465 (0.344)	1.219 (0.360)	1.887 (0.363)	0.810 (0.309)
LS Mean Diff (SE)	N/A	-1.414 (0.325)	-0.661 (0.344)	N/A	**-1.076** (0.289)
95% CI of LS Mean Diff	N/A	(-2.054, -0.775)	(-1.337, 0.015)	N/A	(-1.645, -0.508)
p-value	N/A	<.0001	**0.0552**	N/A	0.0002

LS = least squares; SE = standard error; N/A = not applicable .
N = The total number of subjects in each group under the SIGMAR1/COL24A1 WT Homozygous population. n = Number of subjects with non-missing data for the specific Category/Parameter and model covariates.
[1] Mixed Models for Repeated Measures (MMRM) model is used and include treatment group (one model include all treatments in individual group comparison, the other include treatments for combined group comparison), visit, country, SIGMAR1 gene variant status, and treatment group by visit interaction as fixed effects, the baseline of CDR-SB Total Score, natural log of baseline Nf-L, whether receiving concomitant AD medication at baseline, baseline MMSE total score (>20 cutoff) as covariate based on Toeplitz covariance structure matrix and Kenward-Roger corrected degree of freedom.

2.6.5.4. Analysis performed across trials (pooled analyses and meta-analysis)

2.6.5.4.1. Indirect comparison with the ADNI Cohort

In an effort to support the claim of presence of a clinically meaningful benefit arising in the long term, the Applicant presented an indirect comparison between the ADAS-Cog13 trajectory from study AD-004 and its long-term extension AD-EP-004 and patients in the ADNI cohort, reportedly "matched for cognitive impairment, age, sex and ApoE4 status, in this order of priority" (see table and figure below).

Figure 7. Indirect comparison with the ADNI cohort

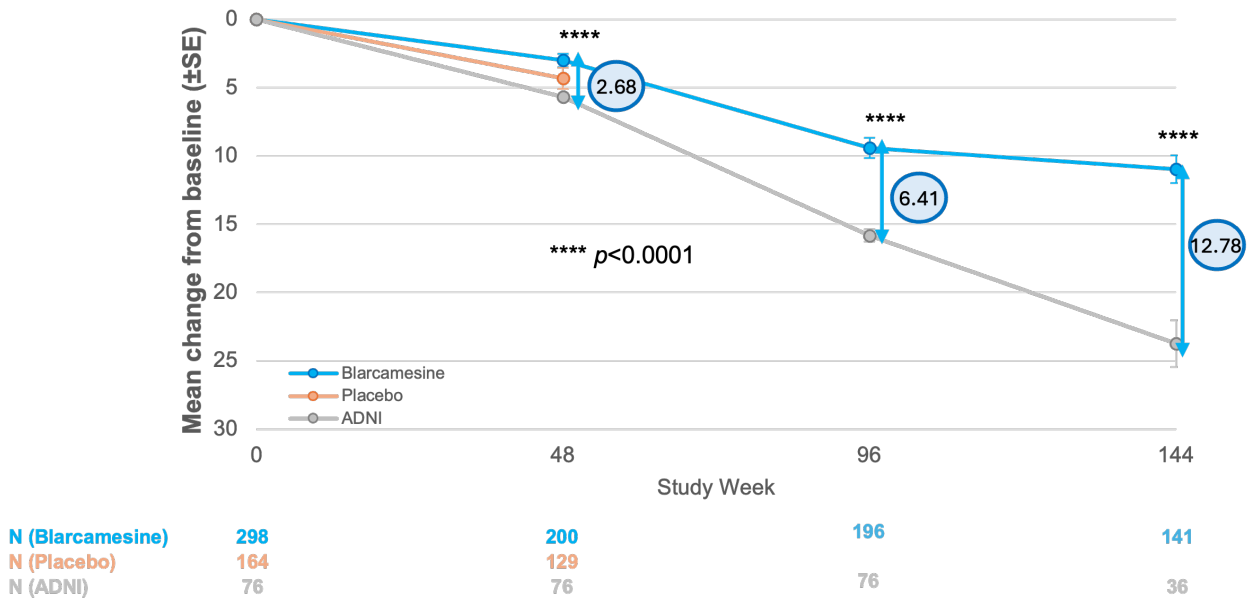


Table 24. ADAS-Cog13 total score, change from baseline, DB/OLE ITT population and ADNI matched control

Week	Treatment Group	Statistics		
		n	Mean Change (SE)	p value vs. ADNI
Week 48	Blarcamesine	200	2.98 (0.490)	<0.0001
	Placebo	129	4.29 (0.774)	0.0853
	ADNI Matched Control	76	5.68 (0.198)	--
Week 96 (OLE Week 48)	Blarcamesine	196	9.41 (0.748)	<0.0001
	ADNI Matched Control	76	15.82 (0.461)	--
Week 144 (OLE Week 96)	Blarcamesine	141	10.97 (0.999)	<0.0001
	ADNI Matched Control	36	23.75 (1.707)	--

2.6.5.5. Supportive study(ies)

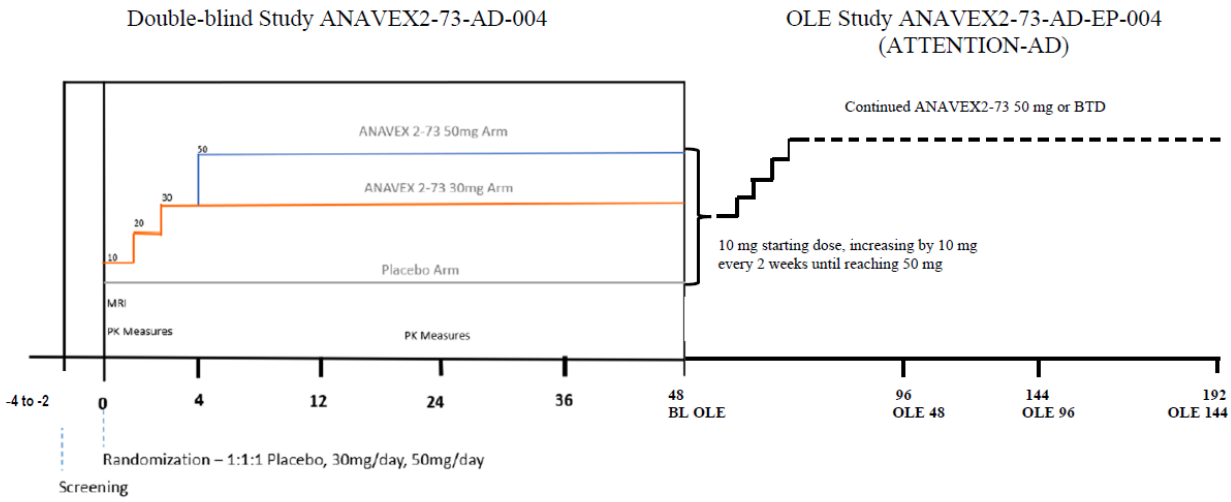
2.6.5.5.1. ANAVEX2-73-AD-EP-004 (ATTENTION-AD)

This was an OLE study following the 48-week ANAVEX2-73-AD-004 double-blind clinical study with an open-label treatment duration of 96 weeks for each participant to evaluate the safety and tolerability of blarcamesine and long-term effects of blarcamesine on cognition and function in participants with early AD. Secondary outcome measures included measures of quality of life (QoL).

The duration of study participation in the OLE was up to 96 weeks for participants in Germany, the Netherlands, and Canada, up to 98 weeks in the United Kingdom (including a 2-week safety follow up visit), and up to 144 weeks for participants in Australia, since the preceding double-blind study (ANAVEX2-73-AD-004) had started in Australia before sites in the other regions (Europe and North

America) were added. The Sponsor terminated the study on 21 May 2024 to facilitate reporting of the results. This did not allow time for the other regions to also participate in the additional OLE extension beyond the initial 96-week OLE period, as done in Australia (144 weeks).

Figure 8. Study designs of the double-blind study and the OLE study



Abbreviations: BL = baseline; BTD = best tolerated dose; OLE = open-label extension

The study was limited to participants who completed the 48-week ANAVEX2-73-AD-004 double-blind clinical study. Some participants received placebo in the double-blind study and were not previously exposed to blarcamesine. The dosing schedule was as follows:

Up-Titration over 10 weeks and then maintenance at up to 50 mg daily or BTD.

- Titration (Weeks 0-10): All participants regardless of their prior dose during the ANAVEX2-73-AD-004 double-blind study underwent the following titration schedule: Treatment began at 10 mg/day blarcamesine for the first two weeks (Week 0 and Week 1), then increased by 10 mg every two weeks over a 10-week period to a maximum maintenance daily dose of up to 50 mg or BTD.
- Maintenance Weeks 11-96/Maintenance for participants in Australia Weeks 11-144: Participants took up to 50 mg blarcamesine per day or BTD.

According to the applicant, baseline disease characteristics were generally consistent across the treatment groups (Continued blarcamesine, Placebo to blarcamesine) for both the OLE Safety and OLE ITT Populations

According to the applicant, the delayed-start analysis for ADAS-Cog13 showed a significant difference between early start and late start treatment groups at Week 144 (LS mean difference -2.70 [95%CI -5.21, -0.19]; P = 0.0348), favouring the early start group. The result at Week 144 demonstrates a larger difference between treatment groups than was observed between blarcamesine and placebo groups at Week 48/EOS in the DB phase (LS mean difference -2.00 [95%CI -3.82, -0.19]; P = 0.0306). At Week 192 (Australia) this observed treatment difference continued to increase (LS mean difference -3.83 [95%CI -6.96, -0.70]; P = 0.0165).

Table 25. Change from DB baseline in ADAS-Cog13 (OLE ITT population and DB ITT and OLE ITT populations) – study ANAVEX2-73-AD-EP-004

Visit Parameter	OLE ITT Population		DB ITT and OLE ITT	
	Placebo to Blarcamesine N = 99	Continued Blarcamesine N = 158	Placebo to Blarcamesine N = 164	Continued Blarcamesine N = 298
Double-blind Week 12				
n	94	149	147	241
LS Mean (SE)	0.45 (1.485)	1.36 (1.437)	1.12 (1.003)	1.87 (0.924)
LS Mean difference (SE)	N/A	0.91 (1.130)	N/A	0.74 (0.861)
95% CI of LS Mean difference	N/A	(-1.31, 3.13)	N/A	(-0.95, 2.43)
p-value for LS Mean difference	N/A	0.4202	N/A	0.3883
Double-blind Week 24				
n	83	148	127	208
LS Mean (SE)	1.55 (1.497)	0.88 (1.436)	1.96 (1.022)	1.42 (0.939)
LS Mean difference (SE)	N/A	-0.67 (1.147)	N/A	-0.54 (0.893)
95% CI of LS Mean difference	N/A	(-2.92, 1.58)	N/A	(-2.30, 1.21)
p-value for LS Mean difference	N/A	0.5593	N/A	0.5423
Double-blind Week 36				
n	88	144	122	187
LS Mean (SE)	2.03 (1.494)	1.64 (1.438)	2.98 (1.034)	2.54 (0.956)
LS Mean difference (SE)	N/A	-0.39 (1.141)	N/A	-0.44 (0.915)
95% CI of LS Mean difference	N/A	(-2.63, 1.85)	N/A	(-2.24, 1.35)
p-value for LS Mean difference	N/A	0.7322	N/A	0.6296
Double-blind Week 48				
n	91	151	122	191
LS Mean (SE)	4.25 (1.489)	2.72 (1.435)	5.33 (1.041)	3.32 (0.965)
LS Mean difference (SE)	N/A	-1.53 (1.133)	N/A	-2.00 (0.925)
95% CI of LS Mean difference	N/A	(-3.75, 0.70)	N/A	(-3.82, -0.19)
p-value for LS Mean difference	N/A	0.1783	N/A	0.0306
Week 96 (OLE Week 48)				
n	64	126	64	126
LS Mean (SE)	9.51 (1.546)	9.86 (1.453)	10.34 (1.177)	10.47 (1.029)
LS Mean difference (SE)	N/A	0.35 (1.219)	N/A	0.13 (1.104)
95% CI of LS Mean difference	N/A	(-2.05, 2.74)	N/A	(-2.04, 2.29)
p-value for LS Mean difference	N/A	0.7764	N/A	0.9066
Week 144 (OLE Week 96)				
n	47	92	47	92
LS Mean (SE)	**14.75** (1.631)	**12.34** (1.490)	15.49 (1.300)	12.79 (1.099)
LS Mean difference (SE)	N/A	**-2.42** (1.359)	N/A	**-2.70** (1.278)
95% CI of LS Mean difference	N/A	(-5.09, 0.25)	N/A	(-5.21, -0.19)
p-value for LS Mean difference	N/A	**0.0759**	N/A	**0.0348**
Week 192 (OLE Week 144) (Australia)				
n	30	54	30	54
LS Mean (SE)	19.31 (1.812)	15.69 (1.612)	20.02 (1.515)	16.19 (1.258)
LS Mean difference (SE)	N/A	**-3.63** (1.652)	N/A	**-3.83** (1.595)
95% CI of LS Mean difference	N/A	(-6.87, -0.38)	N/A	(-6.96, -0.70)
p-value for LS Mean difference	N/A	**0.0285**	N/A	**0.0165**

Abbreviations: ADAS-Cog13=Alzheimer Disease Assessment Scale-Cognition; CI=confidence interval; DB=double blind; ITT=intent-to-treat; LS=least squares; max=maximum; min=minimum; MMRM=Mixed Models for Repeated Measures; OLE=Open-Label Extension; SE=standard error

Note: Baseline is baseline in ANAVEX2-73-AD-004 study. Week 192 (OLE Week 144) was for participants in Australia.

The delayed-start analysis for ADCS-ADL showed similar trends: the treatment difference between early start and late start groups at Week 144 (LS mean difference 2.32 [95%CI -0.64, 5.28]; P =0.1250) while non-significant, was greater than at Week 48 of the DB study between blarcamesine and placebo groups (LS mean difference 0.89 [95%CI -1.28, 3.06]; P = 0.4214). This trend continued at Week 192 (Australia), with the difference between early and late start groups reaching statistical significance (LS mean difference 4.30 [95%CI 0.66, 7.94]; P = 0.0206).

Table 26. Change from DB baseline in ADCS-ADL (OLE ITT population and DB ITT and OLE ITT populations) – study ANAVEX2-73-AD-EP-004

Visit Parameter	OLE ITT Population		DB ITT and OLE ITT	
	Placebo to blarcamesine N = 99	Continued blarcamesine N = 158	Placebo to blarcamesine N = 164	Continued blarcamesine N = 298
Double-blind Week 48				
n	92	152	126	194
LS Mean (SE)	-4.15 (1.737)	-4.05 (1.682)	-7.32 (1.220)	-6.43 (1.139)
LS Mean difference (SE)	N/A	0.10 (1.360)	N/A	0.89 (1.108)
95% CI of LS Mean difference	N/A	(-2.57, 2.77)	N/A	(-1.28, 3.06)
p-value for LS Mean difference	N/A	0.9408	N/A	0.4214
Week 96 (OLE Week 48)				
n	70	132	70	132
LS Mean (SE)	-13.80 (1.796)	-11.80 (1.700)	-16.38 (1.375)	-13.85 (1.217)
LS Mean difference (SE)	N/A	2.00 (1.441)	N/A	2.54 (1.303)
95% CI of LS Mean difference	N/A	(-0.83, 4.83)	N/A	(-0.02, 5.09)
p-value for LS Mean difference	N/A	0.1660	N/A	0.0519
Week 144 (OLE Week 96)				
n	50	106	50	106
LS Mean (SE)	**-19.90** (1.911)	**-17.98** (1.735)	-22.08 (1.541)	-19.77 (1.291)
LS Mean difference (SE)	N/A	**1.92** (1.601)	N/A	**2.32** (1.509)
95% CI of LS Mean difference	N/A	(-1.22, 5.06)	N/A	(-0.64, 5.28)
p-value for LS Mean difference	N/A	**0.2305**	N/A	**0.1250**
Week 192 (OLE Week 144) (Australia)				
n	34	64	33	64
LS Mean (SE)	-25.87 (2.127)	-21.91 (1.867)	-27.75 (1.791)	-23.44 (1.461)
LS Mean difference (SE)	N/A	3.96 (1.922)	N/A	**4.30** (1.855)
95% CI of LS Mean difference	N/A	(0.19, 7.73)	N/A	(0.66, 7.94)
p-value for LS Mean difference	N/A	0.0396	N/A	**0.0206**

2.6.5.5.2. ANAVEX2-73-002

A phase IIa study of Anavex2-73 adaptive-trial-design with repeated doses, maximum tolerated dose finding, pharmacodynamic investigation and bioavailability evaluation was conducted in patients with mild to moderate Alzheimer's disease with a 52 week open label follow-up period.

This was a Phase 2a study consisting of two parts (Part A and Part B).

The first part (Part A) was a sequentially randomized, open-label, cross-over, adaptive design study lasting approximately 36 days; this part was conducted at a Phase 1 Unit. The second part (Part B) was an open-label extension for an additional period of 52 weeks of treatment with oral ANAVEX2-73, so as to establish a longer drug effect for the subjects who chose to continue on an oral daily dose; this part was conducted at 3 clinical sites.

The first part (Part A) of the study included 2 periods: in the first period either intravenous (IV) or oral formulation of ANAVEX2-73 was administered and in the second period the oral or intravenous (IV) form of ANAVEX2-73 was administered in a crossover design. The first period (Period 1) included 12 administrations (either oral or IV) and the second period (Period 2) included 11 administrations (either oral or IV).

All subjects had the option to continue taking study drug in the extended open-label study for assessment of longer cognitive effects for an additional 52 weeks (Part B). The subjects who received the IV dose form during the second period of Part A were switched over to a corresponding oral dose in Part B and subjects who received the oral dose continued on the same oral dose.

The planned doses in Part A were 3 and 5 mg for IV formulation and 30 and 50 mg for the oral formulation. However, the adaptive design of the study allowed for dose adjustments based on PK and safety results; therefore, doses ranging from 10 mg to 60 mg for the oral formulation and 1 mg to 20 mg for the IV formulation were possible.

Schedule of visits was Screening, Day 1, Day 25, Day 36 (Part A)/Week 1 (Part B) in Part A and Weeks 12, 26, 36, and 48 in Part B.

Number of patients (planned and analysed):

In Part A, a total of 32 subjects were planned for enrolment. Thirty-two subjects participated in Part A and 30 subjects participated in Part B. The Safety (Full Analysis) and PK analysis populations included 32 subjects, and the Evaluable Efficacy Population included 30 subjects.

Part A was approximately 36 days (Period 1 and Period 2 with a wash-out period between Period 1 and Period 2) and Part B was 52 weeks.

Test product, dose and mode of administration, batch number.

Two capsule strengths (10 mg and 20 mg) were manufactured to cover the planned oral dose ranges (10 mg to 60 mg). The infusion formulation of the drug substance consisted. The batch numbers will be included in the final report.

Exploratory Efficacy:

Subjects with higher MMSE baseline (> 18) had higher chances of maintaining or improving their cognitive state at week 48 compared to subjects with lower baseline MMSE.

Overall, according to the applicant, the ADCS-ADL AAUC analysis indicated that while improvements (>100% of AAUC baseline score) were not observed in most subjects, the AAUC score was typically stable over 48 weeks. Furthermore, more than 90% of subjects maintained at least 85% of the ADCS-ADL AAUC baseline score.

The covariate analysis showed the baseline may affect detection task (DET) but not other CBB+ISL metrics and suggested subjects with slower detection speed at baseline may have more room to improve than subjects with quicker detection speed at baseline. The most responses were observed for the One Card Learning and One Back in CBB+ISL metrics, with 11 of 24 subjects meeting the responder criterion at Week 48. Because the study was an exploratory one, a responder criterion was created to enable the extraction of hidden knowledge from the study data. The responder criterion was at least a 5% improvement in corresponding speed relative metric score compared to baseline value and at least 2 or more correct responses than baseline line correct responses for count related metrics The applicant is of the view that overall, there were 75% (n=18) of subjects responded in at least one or more metrics.

There were serendipitous benefit occurrences for 8 subjects including improvement in mood, cognition and memory based on input from patients and care takers.

2.6.5.5.3. ANAVEX2-73-003

Study 73-003 is an extension study in patients with mild to moderate Alzheimer's Disease who had previously participated in Study ANAVEX2-73-002.

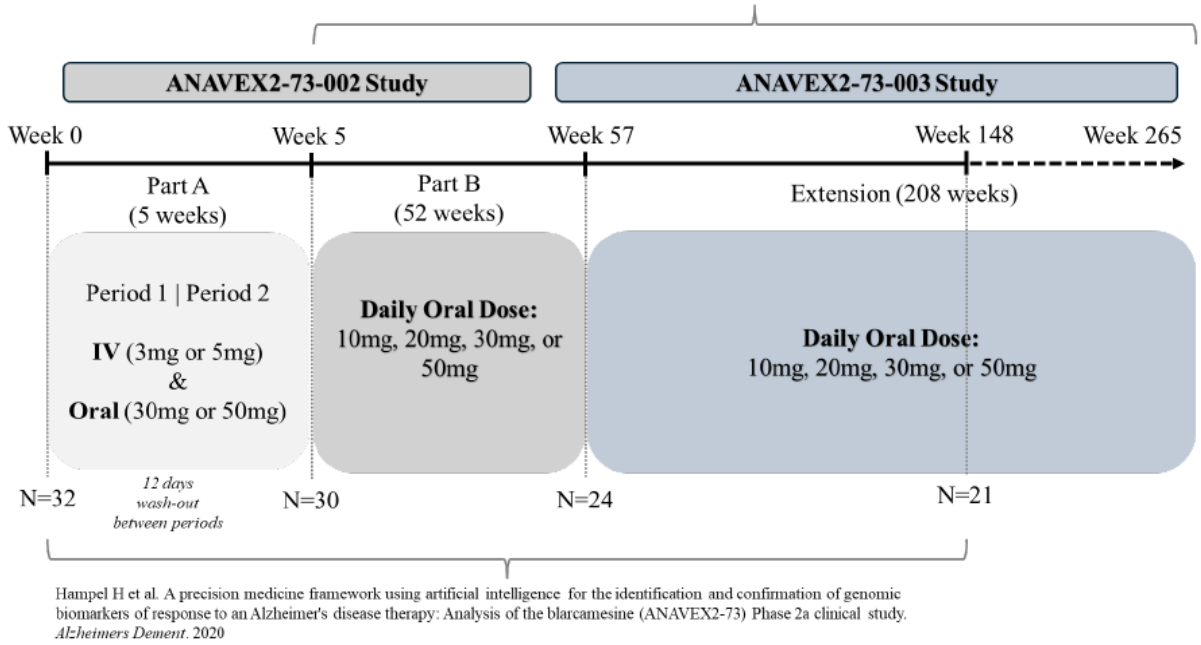
The primary objective of this study was to evaluate the long-term safety and tolerability of blarcamesine. The secondary efficacy objectives of this study were to evaluate the long-term efficacy of blarcamesine in the Mini-Mental State Examination (MMSE), Alzheimer's Disease Cooperative Study

- Activities of Daily Living (ADCS-ADL) and Hamilton Psychiatric Rating Scale for Depression (HAM-D).

This open-label extension (OLE) study was originally designed to provide up to 104 additional weeks of treatment with blarcamesine to eligible participants with mild to moderate Alzheimer's disease (AD) who had previously completed Anavex-sponsored study ANAVEX2-73-002 and who wanted to continue receiving treatment with blarcamesine; the study was subsequently amended to allow participants to continue for an additional 104 weeks (for a 208-week total duration in this extension study). In Study ANAVEX2-73-002, participants who completed Part A and Part B were exposed to blarcamesine for a total of 57 weeks. The results of Study ANAVEX2-73-002 (Part A and Part B) have been reported in a published CSR (ANAVEX2-73-002).

This study was limited to participants who completed Part B of the Phase 2a study, ANAVEX2-73-002. The QD oral doses of blarcamesine tested in this study ranged from 10 mg to 50 mg and were initially based on the dose the participant received in Part B of study ANAVEX2-73-002

Figure 9. Synopsis part A and part B of Open-Label Study ANAVEX2-73-002 (57 weeks) and Open-Label Extension ANAVEX2-73-003 (208 weeks)



Hampel H et al. A precision medicine framework using artificial intelligence for the identification and confirmation of genomic biomarkers of response to an Alzheimer's disease therapy: Analysis of the blarcamesine (ANAVEX2-73) Phase 2a clinical study. *Alzheimers Dement*. 2020

The original Study ANAVEX2-73-002 (published CSR ANAVEX2-73-002) consisted of Part A and Part B (57 Weeks).

Of the 25 participants who completed Part B of the index study (ANAVEX2-73-002), 24 participants were enrolled in the OLE (10 mg, N=8; 20 mg, N=8; 30 mg, N=7; 50 mg, N=1) and all 24 were included in the Full Analysis Population, which was defined as all enrolled participants who received at least 1 dose of study drug in the OLE and who had at least 1 post dose value in any of the 3 efficacy endpoint evaluations in the OLE.

When efficacy measures were assessed by dose group, no clear dose effects were observed, which was not unexpected given the small sample size.

The MMSE LS means in the high concentration and low/medium concentration groups began to separate at ~Week 91 in the OLE and this difference was sustained over the course of treatment,

according to the applicant, although it did not reach statistical significance. The high concentration group showed a LS mean (standard error [SE]) change from baseline of -6.0 (2.95) compared to a LS mean change of -11.1 (2.77) in the low/medium concentration group after 208 weeks of treatment with blarcamesine during the OLE, indicating less worsening in dementia symptom severity in participants in the high concentration group compared with the medium/low concentration group.

With regard to the ADCS-ADL, LS means for the high and low/medium concentrations groups began to separate at ~Week 26 of the OLE with the difference achieving statistical significance beginning at Week 65 (of the OLE), according to the applicant. The LS mean difference between concentration groups in total ADCS-ADL score remained statistically significant at most subsequent visits throughout the remainder of the OLE treatment period and was statistically significant overall (P = 0.035). After 208 weeks of treatment with blarcamesine in the OLE, the LS mean (SE) change from baseline was - 22.1 (7.34) in the high concentration group compared with - 48.5 (6.56) in the low/medium concentration group (P = 0.009). This difference between groups could indicate less decline in ability to perform activities of daily living in participants in the high concentration group compared with the medium/low concentration group.

2.6.6. Discussion on clinical efficacy

Blarcamesine (10 mg capsules) is intended as a disease modifying oral therapy for the treatment of Alzheimer´s disease.

The proposed indication after the submission of the applicant's latest responses to CHMP was:

> "Blarcamesine is indicated as add-on therapy for the treatment of early Alzheimer's disease in adults with mild cognitive impairment (MCI) due to Alzheimer's disease or early-stage mild dementia due to Alzheimer's disease, in patients with SIGMAR1 wild-type genotype."

The attempt to demonstrate efficacy is based on a single pivotal trial. Even with a successful trial, this would have been a problematic approach due to the need of replication, especially for a new mechanism of action. However, the trial should be considered formally negative as one of the co-primary endpoint failed to achieve statistical significance, for a number of late changes to the analysis methods and lack of full and detailed pre-specification of the analyses.

As the indication refers to a subgroup, it firstly bears noting that the results in a subgroup can only be exploratory, lacking statistical significance in the overall population and lacking replication. Secondly, the results in the SIGMAR1 wild-type genotype (with two sets submitted) are not markedly different from the overall population.

The applicant also presented another subgroup (not corresponding to the indication sought), of patients who are both SIGMAR1 and COL24A1 WT homozygotes. This is however not interpretable due to the lack of pre-specification.

Design and conduct of clinical studies

The inclusion criteria are considered acceptable, and the applicant's choice of two co-primary endpoints (ADAS-Cog13 and ADCS-ADL) is also in principle acceptable. It was specified originally that the study will be successful if both primary endpoints reach statistical significance. While acknowledging that the study was designed prior to recent guideline recommendations, the deviation from the success criterion defined cannot be accepted a posteriori.

There are also a number of issues with respect to pre-specification of the analysis methods (dichotomised versus continuous endpoint, selection of covariates, covariance structure) and of the multiplicity control that support the notion that the results cannot be regarded as truly significant. The applicant's claim that who drafted the SAP lacked experience in Alzheimer's disease trial design and analysis does not increase the credibility of the findings. The approach for analysing the data was changed after the end of the study. The applicant openly admits problems with an unexperienced statistical analysis team. However, changes to the SAP are not acceptable after the data are seen and post-hoc analyses in knowledge of the data cannot render a failed study successful.

The sample size (N=462 patients in the ITT population) is considered limited compared to the millions of Alzheimer's disease patients in Europe. Furthermore, the planning assumption that there will be a 33% dropout rate turned out not to be correct in the case of blarcamesine. The percentages of patients discontinuing due to adverse events were higher in the blarcamesine groups 30mg and 50 mg compared to the placebo group. This was the case for both the early discontinuations and the ones after week 12. Only 53.3% of the participants in the 50mg blarcamesine group, completed the study. In the case of placebo, 80% of the participants completed the study.

Efficacy data and additional analyses

The initial number of patients randomised n=508 (the ITT population included 298 patients in the blarcamesine active group) is considered very limited compared to the number of Alzheimer's patients worldwide. It is estimated that 7 million people in Europe alone already live with Alzheimer's disease. With a rapidly ageing population, it is a growing public health concern worldwide as this number is projected to rise to a staggering 14 million by 2030 (https://www.braincouncil.eu/projects/rethinking-alzheimers-disease/#:~:text=About%20Alzheimer's%20Disease&text=It%20is%20estimated%20that%207,staggering%2014%20million%20by%202030). An estimated 6.9 million Americans age 65 and older are living with Alzheimer's dementia in 2024. Of the total U.S. population: about 1 in 9 people (10.9%) age 65 and older has Alzheimer's dementia (https://www.alz.org/getmedia/76e51bb6-c003-4d84-8019-e0779d8c4e8d/alzheimers-facts-and-figures.pdf).

The participants in study AD-004 were receiving concomitant medication, with 57-64% of them receiving symptomatic Alzheimer's Disease medication. This was not in accordance with the applicant's proposal, since oral blarcamesine was promoted as an alternative monotherapy treatment option for AD. Later in the procedure the applicant modified the proposed indication and the Product Information to suggest that blarcamesine is intended as add-on treatment.

Study Participants

A number of participants were excluded from the analyses, and this was disproportionate in blarcamesine (n=37) and placebo (n=4) groups. The applicant provided the numbers of discontinuations in the two groups. According to the applicant, the majority of discontinuations reported in the active group were related to adverse events, which arose during the initial up-titration phase and were predominantly mild-to-moderate in severity. The applicant claims that since these discontinuations occurred before any post-baseline efficacy was recorded, there is no reason to believe that these discontinuations had an impact on the efficacy results. This is not necessarily the case, as the population becomes selected on post-randomisation events, and especially since the number of discontinuations was high. It is also noted that the definition of ITT population does not correspond to the ITT principle and was changed in one of the protocol amendments. These issues further lower the confidence in interpreting trends from this negative study.

The exclusion of patients and more broadly the handling of missing data was not considered

satisfactory as not conservative. The applicant provided the requested analyses using Pattern Mixture Model with Control-based Pattern Imputation. Once this method is applied, also the ADAS-Cog13 results are negative in the ITT population at Week 48 for the pre-specified dose groups (p=0.0964 and p=0.1406 for blarcamesine 30 mg and 50 mg groups, respectively) and for the post hoc pooled dose group (p=0.0676). For the SIGMAR1 WT Homozygous, results are nominally significant for the low-dose group (p = 0.0370) and for the pooled dose (p = 0.0237) but not for the high dose group (p = 0.0746).

Discontinuations

Subjects who were unable to tolerate the lowest dose of 10 mg were discontinued from the Study. Moreover, participants who experienced any dose interruption of >10 consecutive days were withdrawn from the study.

In total 170 (33.5%) participants discontinued early from the study. The majority of patients discontinued due to AEs (70%). Withdrawal of consent was the second most frequent reason (15.3%) followed by protocol violations (4.7%). One participant in the 50 mg/day group discontinued due to death. Early terminations due to AEs in the active treatment groups occurred predominantly before Week 24. However, this is considered questionable, if discontinuations before Week 24 can still be considered "early discontinuations".

The discontinuations up to Week 24 were numerically higher in the 50 mg dose compared to the 30 mg dose group and the cause of discontinuation due to AEs was numerically higher in the 50 mg dose group compared to the 30 mg dose group up to Week 12. These data concern mainly safety (see below) but also make the efficacy results difficult to interpret, in conjunction with data missingness and handling of missing data.

Dosage Regimen

It is noted that the primary analysis was comparing combined 30 mg and 50 mg groups vs placebo in primary, secondary and exploratory endpoints as specified in the SAP. As the SAP was largely not followed, it also bears noting that other features of the protocols (e.g. the sample size calculation) seemed to refer to separate primary comparisons of each dose against placebo. The applicant justifies combining the two dose groups with the similarities of the average daily exposure. Even if it had been fully pre-specified (which is not the case), this pooling approach would not be recommended in this single pivotal clinical trial. In addition to methodological issues, there are also tolerability issues in the 50 mg group. Taking into account the worse safety profile of the higher dose and generally comparable efficacy results for both doses, such a way of presenting the data is not considered optimal. The 30 mg daily dose was found as the best tolerable maintenance dose, and this was proposed by the applicant in the SmPC. Hence, results for patients assigned to that target dose should have been considered primarily, but were not.

Duration of study

The duration of the study was too short, for an early AD population to show a treatment effect, only 48 weeks, whilst the recommended minimum duration for a disease modifying treatment is 18 months in the EMA Guideline (CPMP/EWP/553/95 Rev.2, 22 February 2018). The applicant acknowledged the guideline recommendation for longer trial durations in early AD and committed to conducting a longer-term confirmatory study (≥18 months) post-approval to validate durability of effect and further establish disease modification. The commitment of the applicant to conduct one longer-term study post-approval is noted. However, a CMA cannot be granted unless a favourable benefit/risk ratio for blarcamesine is shown (see below).

Results in the ITT population

One of the co-primary endpoints, ADAS-Cog13, showed statistically significant results. Similarly, the secondary endpoint, CDR-SB, showed nominally statistically significant results. However, the functional endpoint, ADCS-ADL, which was the other co-primary endpoint in the pivotal study AD-004 did not reach statistical significance, rendering the study failed (even without considering the other important limitations).

The trial should be evaluated against the appropriate success criterion: both co-primary endpoints had to be significant at two-sided 0.05 alpha level. When evaluated against this standard, the trial is negative. The Applicant seemed to refer in some of their responses to CHMP to a post-hoc alternative success criterion, whereby at least one of three endpoints (namely the two co-primary and CDR-sb) would need to be significant at a Bonferroni-corrected alpha level. While the Bonferroni correction is known to be conservative when properly applied, this post-hoc definition of success criterion cannot be considered to bear any evidential weight, primarily due to its post-hoc nature.

The applicant argues that the ADCS-ADL questionnaire may not be adequately sensitive for patients with early AD. The design of the study AD-004 and the inclusion of this co-primary endpoint occurred in 2017, before the release of the EMA Guideline on medicinal products for the treatment of Alzheimer's disease and other dementias, in 2018. However, it was the applicant's decision to select the co-primary endpoints and the applicant's responsibility to include an adequate population of patients in the study. Change of the endpoint after finalising the study is not considered acceptable. It is also noted that scientific advice from EMA or any other National Competent Authority for the development in AD was never sought.

Given the discrepancies between the analysis described in the SAP, the analysis described in the protocol, and the analysis reported in the CSR, no analysis can really be considered fully pre-specified. This creates insurmountable limitations to the credibility and interpretability of any claimed positive trend or result.

The Toeplitz covariance structured was not pre-specified and it appears to lower the p-value in the analyses where both it and the unstructured covariance matrix are used. In a late submission during the procedure, the Applicant argues that "[t]he covariance structure and model specification were not post-hoc, but rather selected using standard statistical model selection criteria". Firstly, full pre-specification of the primary analysis method is required, unless algorithms that select features of the analysis methods are demonstrated to preserve type I error. Secondly, in the same submission the applicant points to a paragraph in an annex to the CSR that starts by stating that "The primary analysis of ADAS-Cog13 […] was conducted with an unstructured (UN) matrix as a variance-covariance matrix. […]". Thirdly, given the degree to which the SAP has not been followed in the analysis, even if the algorithm for selecting the covariance structure were precisely defined there a priori, this could hardly be considered pre-specification.

In response to questions on the lack of pre-specification of the covariate selection, the applicant provided analyses excluding ln(Nf-L) and RSGENE covariates, an analysis somewhat closer to the study protocol, since the study protocol did not mention the adjustment for these covariates (neither the SAP did). In this analysis, in none of the dose groups not even a nominal significance was seen for ADAS-Cog13 anymore (30mg: p=0.1043, 50mg: p= 0.0685). This analysis used the Toeplitz covariance structure, that was not pre-specified. The analysis which omits the additional covariates is expected to yield even higher p-values if the unstructured covariance matrix is used. Hence, even if ADAS-Cog13 had been specified as the sole primary endpoint, no significant result would be expected for either of the dose groups.

The lack of nominally significant results on quality of life and caregivers' burden is also noted, and further supports the notion that the trial fails to demonstrate a benefit of treatment.

The definition of ITT, the amount of missing data and its handling also raise concern. While the Applicant has presented a tipping point analysis, this is also affected by selection of patients based on post-baseline attendance to at least one visit. Furthermore, the value by which active group participants would need to shift to reach the tipping point is not completely extreme (e.g. as compared to the treatment effect estimated in the primary analysis), so that this analysis does not support the robustness of the result.

In addition, there were baseline imbalances. It is acknowledged that the applicant has used change-from-baseline and included baseline values as a co-variate in the analysis.

The indirect comparison presented against the ADNI cohort has been reported with insufficient level of detail and it is not interpretable lacking a comprehensive demonstration of the robustness of the matching.

Lastly, the results on biomarker are noted. While not entirely consistent, some of the results might be of exploratory interest, but none of the biomarkers tested – nor the totality of biomarkers tested – can be considered a surrogate for the demonstration of clinical benefit.

Analyses in the SIGMAR1 WT subpopulation

With responses to questions, the applicant focused their application on the SIGMAR1 WT (wild-type) genotype. The applicant stated that the SIGMAR1 WT (wild-type) analysis was pre-specified in the study protocol, based on genetic data from the preceding Phase 2a (ANAVEX2-73-002/003) trials of blarcamesine in Alzheimer's disease and blarcamesine's mechanism of action as a SIGMAR1 agonist. In the statistical analysis plan document, "subgroup analyses based on rs1800866 gene variants" are specified, and the "rs1800866 gene populations" are listed as RS-Negative Gene population (understood to correspond to the SIGMAR1 WT population and the RS Homozygous negative population, also including carriers of one mutated variant). The expected direction of any difference in the subgroup analysis does not appear clearly specified.

However, even for subgroups that were pre-specified, the subgroup analysis remains exploratory and hypothesis generating. Consistent with the EMA Guideline on the investigation of subgroups in confirmatory clinical trials (EMA/CHMP/539146/2013), efficacy cannot be established through restriction to a subgroup following a failure of the primary analysis, even if the subgroup is pre-defined. Confirmation of efficacy necessitates the conduct of one or more additional, appropriately designed clinical trials in the subpopulation. The exceptions detailed in the EMA guideline – namely, circumstances where trial replication is not feasible or the trial involved a large patient population (e.g., cardiovascular disease) yielding robust evidence within pre-defined subpopulations – do not apply in this case. This requirement extends to applications for Conditional Marketing Authorisation (CMA): a demonstration of efficacy is a prerequisite for approval (while acknowledging that in the case of a CMA, the evidence provided may not be fully comprehensive).

The results in the SIGMAR1 WT subpopulation for the change from baseline for ADAS-Cog13 and CDR-SB are not nominally statistically significant, even when pooling the results and calculating for the blarcamesine active total, when analysed with the unstructured covariance structure and without covariates.

As discussed above, the presentation of the results by the applicant is confusing, since two separate sets of results are reported: one for the SIGMAR1 WT (not nominally statistically significant using MMRM Model, Unstructured Covariance, No Covariates) and another one for the rs1800866 Genotype: Absence populations (using MMRM Analysis, Toeplitz covariance). However, the two sets include the same patients.

In any case, it should be pointed out that the restriction of the results to a subpopulation in this failed study cannot be accepted, even if the issues around the plausibility of efficacy in the subgroup being larger were solved and if the set of results considered were the ones reported as relating to the "rs1800866 Genotype: Absence".

Post-hoc analyses in the SIGMAR1/COL24A1 WT Homozygous Population

The applicant has also submitted data derived from genetically defined patient population analyses, in the SIGMAR1/COL24A1 wild-type population. The applicant claims that the MoA of blarcamesine (ANAVEX2-73) is based on the agonistic properties for the SIGMAR1/S1R, which lead to the restoration of impaired autophagy. It is postulated that blarcamesine interacts via a specific S1R motif with co-functioning autophagy proteins to promote autophagosome biogenesis, autophagic cargo reception, and lysosome fusion. This mechanism, according to the applicant, aligns with emerging understanding of how the brain's extracellular matrix (ECM) directly influences neural autophagy.

The applicant is of the view that in carriers of the COL24A1 wildtype allele, the preserved extracellular matrix (ECM) structure, mediated by the wildtype COL24A1 protein, likely provides the necessary microenvironmental framework for blarcamesine's mechanism of S1R-mediated autophagy enhancement to function optimally in countering Alzheimer's pathology. COL24A1, encodes the alpha-1 chain of fibrillar collagen XXIV, a potential link between brain ECM composition and blarcamesine response. These assumptions are considered hypothesis generating, since they are not supported by specific non-clinical or clinical pharmacology studies. Moreover, they should be seen in the context of a post-hoc decision. The applicant indeed in its responses acknowledged that COL24A1 was identified post hoc in this study and is therefore exploratory and hypothesis generating. The analysis which "utilized a gene-guided patient selection" is a post-hoc analysis, which is likely to be data driven and with a high risk of bias.

In addition to the considerations on subgroup analyses made above, there are additional limitations:

- There is no external evidence that the proposed SIGMAR1/COL24A1 WT subpopulation is a well-defined and clinically relevant entity. Particularly, the subgroup was not considered when planning the trial, but identified post-hoc based on exploratory analyses
- A pharmacological rationale, or a mechanistically plausible explanation as to why there could be different efficacy in this subpopulation needs to be demonstrated, also with non-clinical and clinical pharmacology studies. The explanations provided by the applicant in this respect can only be considered as hypotheses that are not based on evidence.

Additionally, and while noting that due to the above considerations even very convincing results in this subgroup would in any case not lead to a demonstration of efficacy, several flaws have been identified. The values from the ADAS-Cog13 show a noticeable improvement (with decreased LS mean) at week 24 for the blarcamesine 30 mg group, but a response similar to placebo was observed for the 50 mg group. This is not considered biologically plausible.

The results for the ADCS-ADL do not have a similar shape or pattern compared to the results from ADAS-Cog13. They show a constant deterioration of the patients' condition. The 30 mg and the placebo groups are similar at week 12 whilst the 50 mg is in a worse condition compared to the other two groups. It also appears that the placebo group has a linear deterioration over time, which is not expected. ADNI (Alzheimer's Disease Neuroimaging Initiative) and AIBL (Australian Imaging, Biomarkers and Lifestyle) studies show non-linear cognitive and biomarker progression in MCI or mild AD patients.

The results from the various endpoints do not show a trend towards the same direction. Each of the endpoints show a different pattern and each endpoint is not in accordance with another endpoint. The two treatment groups, blarcamesine 30 mg and 50 mg have different trends depending on the measurement tool used. And the analyses for the placebo group (with the exception of the analysis for the ADAS-Cog13 endpoint) show an almost perfect linear disease progression. This is not biologically plausible and does not correspond with our knowledge of Alzheimer's disease up to now.

An indirect comparison between the ADAS-Cog13 trajectory from study AD-004 and its long-term extension AD-EP-004 and reportedly patients in the ADNI cohort has also submitted Given the lack of detail provided, it is impossible to assert that the analysis supports any efficacy claim. The Applicant reports that "at time point 48 weeks after baseline, end of the double-blind study, the external control group and the placebo-to-blarcamesine group are not significantly different in terms of mean change (p=[0.0853]) in ADAS-Cog13 score". However, the lack of statistical significant difference between the placebo and the matched cohort decline is not a sufficient demonstration that the matching has worked, especially given that the point estimate for the difference in mean change from baseline between the placebo arm and the cohort (1.39) is far from zero, and also similar to the point estimate of the difference between Blarcamesine and Placebo (1.31).

Supportive studies

Patients, who completed the pivotal study could continue the treatment in a 144-week long-term extension Study ANAVEX2-73-AD-EP-004.

Additional data from patients with mild-to-moderate AD have been provided from a randomized, open-label 57-week Study ANAVEX2-73-002 and a 208-week long-term extension Study ANAVEX2-73-003.

ANAVEX2-7373-AD-EP-004 (ATTENTION-AD)

It is noted that a different dosing titration schedule was used in the OLE Study compared to the pivotal study AD-004. It is not clear why the patients in this extension study were re-titrated over a 10-week period to a maximum maintenance daily dose of up to 50 mg. The patients who were on blarcamesine in study AD-004 did not continue with their maintenance dose in the OLE EP-004. None of the patients having continuously been treated with blarcamesine in study -004 and EP-004, entered the extension period without a gap. The mean gap in dosing was ~16 days. The titration scheme used in this study does not reflect the proposed posology.

Study visits were performed every 24 weeks through the end of the study. However, the frequency of visits is not considered optimal. The applicant should have provided a justification for the choice of such visits frequency and discuss if visits every 24 weeks can be considered sufficient to assess adequately the study population.

The primary objective of the study was to assess the safety and tolerability of blarcamesine. The secondary objective was to determine whether blarcamesine modifies cognition, behaviour, or QoL.

It is also unclear why 44 participants in Australia were receiving for 20 weeks blarcamesine in the form of oral solution (5 mg/ml), instead of blarcamesine capsules. The applicant states that this was related to the unavailability of capsules. Moreover, it is noted that unusually high number of those 44 subjects terminated the OLE early and did not complete the treatment (22 subjects).

Of the 338 participants who completed the index study (ANAVEX2-73-AD-004), 300 enrolled into the OLE (121 who received placebo and 179 who received blarcamesine in the index study). Of these, 257 were included in the ITT population, which was defined as any OLE participant who received at least 1 dose of study drug in the OLE, had an OLE baseline assessment, and had at least 1 post-baseline efficacy

assessment in OLE study.

However, of the 300 participants in the OLE study, approximately 43% completed the study and 57% discontinued early. The most common reason for early discontinuation was the occurrence of an AE (44.2% of total participants). This very high incidence of discontinuations in the studies is considered to weigh very heavily on the B/R assessment.

Since the study was open-label, the efficacy results should be treated with caution.

In conclusion, the results from AD-EP-004 cannot be considered supportive of the preliminary results of study AD-004, even in the case that the pivotal study had been successful.

Studies in mild to moderate AD

Study 73-002 and its open label extension Study 73-003 were conducted in participants with mild to moderate AD, a different population than in the pivotal trial AD-004 and its open label extension EP-004. This small dose finding study and its open-label extension cannot provide support for the applied for indication.

ANAVEX2-73-002

A Phase II a Study of ANAVEX2-73 adaptive-trial-design with repeated doses, maximum tolerated dose finding, pharmacodynamic investigation and bioavailability evaluation was conducted in a limited number of patients with mild to moderate Alzheimer's Disease with a 52-week open label follow-up period.

Study AVANEX2-73-AD-002 was conducted in 2 parts (Part A and Part B) in male or female participants aged 55 to 85 years with mild to moderate AD. Part A was a sequentially randomized, open-label, cross-over, adaptive design study lasting approximately 36 days that compared an intravenous (IV) formation to an oral formulation in order to estimate the absolute bioavailability of blarcamesine. Part B was an open-label extension in which participants who completed Part A could receive an additional 52 weeks of treatment with the oral formulation.

The primary objective was to confirm the maximal tolerated dose (MTD) of ANAVEX2-73 in subjects with mild to moderate AD by assessment of safety parameters.

With respect to the efficacy evaluation, statistically significant difference in ADCS-ADL between any two groups was not achieved. Moreover, no differences were observed over time when comparing median ADCS-ADL total score values between visits or between average daily dose groups.

According to the applicant, the ADCS-ADL AAUC analysis indicated that while improvements (>100% of AAUC baseline score) were not observed in most subjects, the AAUC score was typically stable over 48 weeks. Furthermore, more than 90% of subjects maintained at least 85% of the ADCS-ADL AAUC baseline score. There were 75% (n=18) of subjects responded in at least one or more metrics.

Thirty-two subjects participated in Part A and 30 subjects participated in Part B. In study 73-002, definitive conclusions regarding MTD have not been made. A preliminary assessment suggested that the MTD was slightly above the maximum planned dose of 50 mg. Of note, during the course of the procedure, the maximum daily dose in the product information has been changed from 50 mg to 40 mg and further down to 30 mg. Furthermore, no definitive conclusions could be drawn from this study. The main limitations were the small number of subjects in each treatment arm and also dose adjustments were permitted during the study. It is also noted that five subjects (2 in Part A and 3 in Part B) prematurely discontinued study drug due to TEAE(s).

Overall, study ANAVEX2-73-AD-002 did not provide any data to support the efficacy of blarcamesine.

ANAVEX2-73-003

Study 73-003 is an extension study in patients with mild to moderate Alzheimer's Disease who had previously participated in Study ANAVEX2-73-002 with a primary objective to evaluate the long-term safety and tolerability of blarcamesine.

The Evaluable Efficacy Population included 30 participants. Of the 25 participants who completed Part B of the index study (ANAVEX2-73-002), 24 participants were enrolled in the OLE (10 mg, N=8; 20 mg, N=8; 30 mg, N=7; 50 mg, N=1).

The QD oral doses of blarcamesine tested in this study ranged from 10 mg to 50 mg and were initially based on the dose the participant was receiving at the end of Part B of Study ANAVEX2-73-002. Dose modifications (i.e., for safety reasons) were allowed at the discretion of the principal investigator (PI) at the site where the participant received treatment.

Of the 25 participants who completed Part B of the index study (ANAVEX2-73-002), 24 participants were enrolled in the OLE (10 mg, N=8; 20 mg, N=8; 30 mg, N=7; 50 mg, N=1).

The MMSE results did not reach statistical significance. The results for ADCS-ADL were nominally statistically significant between high and low/medium concentration groups. However, these results were generated by 24 participants who were enrolled in four different dose groups 10 mg, N=8; 20 mg, N=8; 30 mg, N=7; 50 mg, N=1.

In both studies 73-002 and 73-003, only exploratory efficacy analyses (MMSE, ADCS-ADL, CBB+ISLT, EEG/ERP) were conducted in a very limited number of patients (n~30, each study), using the Evaluable Efficacy Population (EEP). No confirmatory efficacy data can be drawn from these two open-label studies. Due to very limited number of subjects and open-label design, no conclusions on efficacy can be made. The results from 73-002 and 73-003 can be at best hypothesis generating in patients with mild to moderate AD.

The two patient populations in the double-blind study and its OLE (AD-EP-004) are different from the open-label phase 2a (73-002) and its extension (73-003). The dosing groups between the two sets of studies were also different. Hence the results from the four studies cannot be combined or pooled together and analysed.

2.6.7. Conclusions on the clinical efficacy

Depending on the stage of the procedure, a different indication has been proposed by the applicant and different analyses have been presented: of the ITT population, of the post-hoc analyses of SIGMAR1/COL24A1 WT Homozygous Population and of the SIGMAR1 WT Population, the latter being specified in the documentation of the applicant as additional exploratory analyses.

The data provided does not demonstrate efficacy of blarcamesine in any claimed indication.

The clinical development program is not considered sufficient to support an indication for the treatment of patients with early AD or any other (sub)population. The clinical parts of the dossier contain a number of deficiencies.

In line with the EMA guidance CPMP/EWP/2330/99, a single pivotal study should be compelling with respect to the internal and external validity, clinical relevance, data quality, and internal consistency. The pivotal study ANAVEX2-73-AD-004 failed to meet its co-primary endpoints. The methodological flaws described above are so many that study ANAVEX2-73-AD-004 can only be considered an exploratory study.

The pre-specified analysis based on SIGMAR1 genotype are not well reported. In any case, they would only be exploratory due to the failure to establish efficacy in the overall population and to the lack of

replication. Furthermore, the results in this subpopulation do not strongly differ from those in the overall population. With respect to the results in the SIGMAR1/COL24A1 WT subpopulation, these are post-hoc, with knowledge of the data in a small and highly selected subgroup of AD patients from a failed study and cannot be considered confirmatory. A convincing mechanistic rationale has not been provided.

Moreover, the selection of the dose is not clear, the pivotal study evaluated efficacy and safety of the 30 mg and 50 mg dose, while the average dose in both groups was even below 30 mg. During the several steps of the procedure, different maximum doses have been proposed by the applicant in the posology section of the SmPC. Also importantly, a very high rate of discontinuations impedes the assessment of the B/R of blarcamesine.

2.6.8. Clinical safety

The safety database of blarcamesine in the treatment of adult patients with Alzheimer's disease and dementia is based on the study ANAVEX2-73-AD-004 ("-004"), described above, including 508 patients with Early Alzheimer's disease, and its completed open-label, up to 144-week long-term extension study ANAVEX2-73-AD-EP-004 ("EP-004") in 300 participants from the parent study. Their protocols included a titration scheme to the assigned dose group (i.e. 30 mg/day or 50 mg/day) that was either 2 or 3 weeks (in study -004) or 10 weeks in duration (in study EP-004). Titration in study EP-004 ended up at 50 mg/day or the best tolerated dose (BTD). A maintenance period with a flexible dose regimen followed the titration period in the two studies, when blarcamesine could have been down- and up-titrated based on tolerability.

In addition, supportive safety data are presented from three clinical studies, i.e. from an open-label Phase 2a study in 32 patients with mild to moderate Alzheimer's disease (ANAVEX2-73-002) up to 52 weeks, its open-label extension ANAVEX2-73-003 (24 patients) up to 208 weeks, and a Phase 1 study (ANAVEX2-73-001) in healthy volunteers.

Safety data for blarcamesine are presented study-wise. No pooling has been applied. Safety data in study -004 and EP-004 is also presented separately by treatment phases, i.e. titration and maintenance phases.

Long-term safety derives from study EP-004, while no longitudinal safety data has been presented for patients treated continuously with blarcamesine from study -004 to study EP-004.

In addition, 74 participants, who continued treatment with blarcamesine after completion of the open-label-extension studies -003 and EP-004, receive blarcamesine in the SAS-B Compassionate Use Program in Australia. No safety data has been provided for these patients.

2.6.8.1. Patient exposure

Exposure to dose and overall extent of exposure

ANAVEX2-73-AD-004

The mean duration of exposure to blarcamesine 30 mg/day and 50 mg/day was 249.8 days and 200.9 days, respectively, versus 295.6 days to placebo.

The best tolerated dose (BTD) was between 10 to 30 mg for the 30 mg group and between 10 to 50 mg for the 50 mg group. Due to the flexible dosing design of the study, the 30 mg and 50 mg assigned dosage arms reached similar daily average doses at each study visit (Weeks 12, 24, and 36) and end of study (Week 48).

ANAVEX2-73-AD-EP-004

The mean duration of exposure for the overall OLE Safety population was 607 days, 526.36 days in the placebo to blarcamesine group and 661.51 days in the continued blarcamesine group.

A substantial proportion of patients had up-titration from 10 mg to 50 mg during the titration period (47.3% of total patients; similar for both treatment groups). In the maintenance period, there was a majority of patients with no down-titration from the entry dose (56.7% of total patients), followed by down-titration of 10 mg and 20 mg.

Subject disposition

ANAVEX2-73-AD-004

A total of 508 participants from 52 sites in 5 countries were randomised across the 3 treatments groups. 170 participants discontinued early from the study, 34 patients from placebo and 136 patients from blarcamesine (20% and 40.2%, respectively) with the main reason being adverse events (placebo: 38.2%; blarcamesine: 77.9%), followed by withdrawal of consent (placebo: 20.6%; blarcamesine: 14%). One participant in the 50 mg/day group discontinued due to death (unrelated to treatment). Early terminations due to AEs in the blarcamesine groups occurred mainly before Week 24, and less frequent thereafter. Overall discontinuations up to Week 24 were more frequent in the 50 mg compared to the 30 mg dose group.

ANAVEX2-73-AD-EP-004

A total of 300 participants from study -004 were enrolled in the OLE study; 172 of 300 patients (57.3%) discontinued early. The proportion of participants completing the study was lower for those who initiated treatment with blarcamesine upon entry into the OLE (33.9%) compared with those who continued blarcamesine during the OLE (48.6%). The most common reason for discontinuation was AEs (44.2% of total participants), and the incidence was higher in the placebo to blarcamesine group as compared to the continued blarcamesine group (48.8% vs. 40.2%).

2.6.8.2. Adverse events

In the summary of clinical safety, adverse events are presented study-wise focussing on the pivotal study -004, followed by supportive information from studies EP-004, -003, -002, and -001.

Overview of adverse events

An overview of adverse events in the five clinical studies contributing to the safety profile of blarcamesine is depicted in the table below.

The proportion of participants with at least 1 TEAE and treated with active drug ranged from 83.3% to 100%, versus 76.8% in the placebo group of study -004. Likewise, the range of treatment-related TEAEs in the four studies ranged from 53.3% to 85.4% (30.4% in the placebo group in study -004). The proportion of participants receiving active drug who had at least 1 SAE ranged from 3.1% (study -002, Part A) to 58.3% (study -003). The proportion of participants receiving active drug who had at least 1 TEAE that led to discontinuation of treatment ranged from 6.2% (study -002, Part A) to 32.2% (study -004). There were four TEAEs leading to death during the clinical programme (one of the four deaths occurred in the placebo group). None was rated as related to the study drug.

Table 27. Comparison of treatment-emergent adverse event brief summary results, all studies, safety population

Parameter	Placebo Study 004 N=168 n (%)	Blarcamesine Study 004 N=335 n (%)	Blarcamesine Study EP-004 N=300 n (%)	Blarcamesine Study 003 N=24 n (%)	Blarcamesine Study 002 Part A N=32 n (%)	Blarcamesine Study 002 Part B N=30 n (%)	All Doses[a] Study 001 N=22 n (%)
Any TEAE	129 (76.8)	324 (96.7)	285 (95.0)	24 (100)	29 (90.6)	25 (83.3)	13 (59.1)
Treatment-related TEAE	51 (30.4)	286 (85.4)	204 (68.0)	20 (83.3)	26 (81.2)	16 (53.3)	–[b]
Serious TEAE	17 (10.1)	56 (16.7)	77 (25.7)	14 (58.3)	1 (3.1)	4 (13.3)	0
TEAE leading to treatment discontinuation	12 (7.1)	108 (32.2)	74 (24.7)	3 (12.5)	2 (6.2)	3 (10.0)	0
TEAE resulting in death	1 (0.6)[c]	1 (0.3)	1 (0.3)[c]	1 (4.2)	0	0	0

Study 001 = Study ANAVEX2-73-001; Study 002 = Study ANAVEX2-73-002; Study 003 = Study ANAVEX2-73-003; Study 004 = Study ANAVEX2-73-AD-004; Study EP-004 = Study ANAVEX2-73-AD-EP-004; TEAE = treatment-emergent adverse event.

[a]. Includes placebo.

[b]. Treatment-related TEAEs were tabulated by TEAE, not by participant.

[c]. This participant is counted twice in this table, in Study ANAVEX2-73-AD-004 and in Study ANAVEX2-73-AD-EP-004. The TEAE that ultimately resulted in death during Study ANAVEX2-73-AD-EP-004 was first reported in Study ANAVEX2-73-AD-004.

Common adverse events/ severity of adverse events/ relationship of adverse events

Study ANAVEX2-73-AD-004

Common adverse events by SOCs and PTs in at least >5% of patients are depicted in the Table below.

Table 28. Study ANAVEX2-73-AD-004: summary of TEAEs occurring in at least >5% of participants, titration and maintenance phases combined, safety population

System Organ Class Preferred Term	Placebo N=168 n (%)	Blarcamesine 30 mg N=167 n (%)	Blarcamesine 50 mg N=168 n (%)	Active Total N=335 n (%)
Total number of TEAEs	413	809	873	1682
Participants with at least one TEAE	129 (76.8)	159 (95.2)	165 (98.2)	324 (96.7)
Participants with TEAEs with frequency ≥5%	71 (42.3)	143 (85.6)	148 (88.1)	291 (86.9)
Nervous system disorders	41 (24.4)	108 (64.7)	133 (79.2)	241 (71.9)
Dizziness	15 (8.9)	66 (39.5)	89 (53.0)	155 (46.3)
Balance disorder	3 (1.8)	16 (9.6)	23 (13.7)	39 (11.6)
Headache	8 (4.8)	15 (9.0)	15 (8.9)	30 (9.0)
Lethargy	3 (1.8)	13 (7.8)	13 (7.7)	26 (7.8)
Somnolence	5 (3.0)	15 (9.0)	11 (6.5)	26 (7.8)
Memory impairment	4 (2.4)	3 (1.8)	11 (6.5)	14 (4.2)
Psychiatric disorders	43 (25.6)	93 (55.7)	108 (64.3)	201 (60.0)
Confusional state	5 (3.0)	37 (22.2)	43 (25.6)	80 (23.9)
Anxiety	6 (3.6)	13 (7.8)	20 (11.9)	33 (9.9)
Abnormal dreams	0	12 (7.2)	13 (7.7)	25 (7.5)
Depressed mood	4 (2.4)	10 (6.0)	11 (6.5)	21 (6.3)
Disorientation	0	5 (3.0)	16 (9.5)	21 (6.3)
Nightmare	3 (1.8)	7 (4.2)	9 (5.4)	16 (4.8)
Gastrointestinal disorders	41 (24.4)	44 (26.3)	38 (22.6)	82 (24.5)
Nausea	5 (3.0)	12 (7.2)	7 (4.2)	19 (5.7)
Constipation	9 (5.4)	8 (4.8)	3 (1.8)	11 (3.3)
Diarrhoea	13 (7.7)	14 (8.4)	17 (10.1)	31 (9.3)
General disorders and administration site conditions	12 (7.1)	42 (25.1)	41 (24.4)	83 (24.8)
Fatigue	2 (1.2)	14 (8.4)	14 (8.3)	28 (8.4)
Feeling abnormal	0	9 (5.4)	8 (4.8)	17 (5.1)
Infections and infestations	26 (15.5)	50 (29.9)	25 (14.9)	75 (22.4)
Urinary tract infection	7 (4.2)	11 (6.6)	6 (3.6)	17 (5.1)
Upper respiratory tract infection	2 (1.2)	9 (5.4)	4 (2.4)	13 (3.9)
Injury, poisoning and procedural complications	22 (13.1)	17 (10.2)	15 (8.9)	32 (9.6)
Fall	16 (9.5)	12 (7.2)	10 (6.0)	22 (6.6)
Musculoskeletal and connective tissue disorders	22 (13.1)	19 (11.4)	19 (11.3)	38 (11.3)
Back pain	9 (5.4)	3 (1.8)	7 (4.2)	10 (3.0)

N=sample size of group; n=number of individuals occurring per adverse event category. Percentages are based on N. Active Total: All participants randomized to either blarcamesine 30 mg or blarcamesine 50 mg. Participants are counted once within each level of summarization.

TEAEs in ≥5% of any group for the titration and maintenance phase are presented in Table 29. In general, TEAEs from the Nervous system disorders SOC and from the General disorders and administration site conditions SOC were more frequently reported during the titration period. The incidence of dizziness, balance disorder, nausea, and fatigue for the blarcamesine treatment groups was higher during the titration phase compared to the maintenance phase. The incidence of confusional state was similar between the titration phase and the maintenance phase for both treatment groups.

Table 29. Study ANAVEX2-73-AD-004: TEAEs occurring in at least ≥5% of participants in any group, by treatment phase, safety population

	Titration Phase				Maintenance Phase			
System Organ Class Preferred Term	**Placebo N=168 n (%)**	**Blarcamesine 30 mg N=167 n (%)**	**Blarcamesine 50 mg N=168 n (%)**	**Active Total N=335 n (%)**	**Placebo N=161 n (%)**	**Blarcamesine 30 mg N=148 n (%)**	**Blarcamesine 50 mg N=153 n (%)**	**Active Total N=301 n (%)**
Total number of TEAEs	95	329	356	685	318	480	517	997
Participants with at least one TEAE	53 (31.5)	123 (73.7)	128 (76.2)	251 (74.9)	113 (70.2)	120 (81.1)	128 (83.7)	248 (82.4)
Participants with TEAEs with frequency ≥ 5%	21 (12.5)	99 (59.3)	105 (62.5)	204 (60.9)	54 (33.5	85 (57.4)	100 (65.4)	185 (61.5)
Nervous system disorders	13 (7.7)	80 (47.9)	94 (56.0)	174 (51.9)	34 (21.1)	62 (41.9)	81 (52.9)	143 (47.5)
Dizziness	10 (6.0)	53 (31.7)	67 (39.9)	120 (35.8)	9 (5.6)	28 (18.9)	48 (31.4)	76 (25.2)
Headache	2 (1.2)	7 (4.2)	8 (4.8)	15 (4.5)	6 (3.7)	8 (5.4)	11 (7.2)	19 (6.3)
Balance disorder	1 (0.6)	12 (7.2)	13 (7.7)	25 (7.5)	2 (1.2)	5 (3.4)	11 (7.2)	16 (5.3)
Lethargy	2 (1.2)	9 (5.4)	7 (4.2)	16 (4.8)	1 (0.6)	6 (4.1)	8 (5.2)	14 (4.7)
Psychiatric disorders	11 (6.5)	55 (32.9)	63 (37.5)	118 (35.2)	35 (21.7)	56 (17.8)	78 (51)	134 (44.5)
Confusional state	1 (0.6)	24 (14.4)	24 (14.3)	48 (14.3)	4 (2.5)	16 (10.8)	24 (15.7)	40 (13.3)
Anxiety	0	8 (4.8)	10 (6.0)	18 (5.4)	6 (3.7)	6 (4.1)	11 (7.2)	17 (5.6)
Depressed mood	1 (0.6)	4 (2.4)	6 (3.6)	10 (3.0)	3 (1.9)	8 (5.4)	7 (4.6)	15 (5.0)
Abnormal dreams	0	7 (4.2)	6 (3.6)	13 (3.9)	0	6 (4.1)	8 (5.2)	14 (4.7)
Disorientation	0	4 (2.4)	5 (3.0)	9 (2.7)	0	1 (0.7)	11 (7.2)	12 (4.0)
Nightmare	1 (0.6)	5 (3.0)	5 (3.0)	10 (3.0)	2 (1.2)	2 (1.4)	8 (5.2)	10 (3.3)
Infections and infestations	3 (1.8)	5 (3.0)	7 (4.2)	12 (3.6)	23 (14.3)	46 (31.1)	20 (13.1)	66 (21.9)
Urinary tract infection	1 (0.6)	3 (1.8)	1 (0.6)	4 (1.2)	6 (3.7)	9 (6.1)	5 (3.3)	14 (4.7)
Upper respiratory tract infection	0	1 (0.6)	1 (0.6)	2 (0.6)	2 (1.2)	8 (5.4)	4 (2.6)	12 (4.0)
Gastrointestinal disorders	17 (10.1)	23 (13.8)	18 (10.7)	41 (12.2)	29 (18)	30 (20.3)	23 (15)	53 (17.6)
Constipation	1 (0.6)	5 (3.0)	1 (0.6)	6 (1.8)	4 (2.5)	8 (5.4)	6 (3.9)	14 (4.7)
Nausea	8 (4.8)	8 (4.8)	13 (7.7)	21 (6.3)	6 (3.7)	9 (6.1)	4 (2.6)	13 (4.3)
Injury, poisoning and procedural complications	2 (1.2)	3 (1.8)	3 (1.8)	6 (1.8)	21 (13)	14 (9.5)	12 (7.8)	26 (8.6)
Fall	0	0	1 (0.6)	1 (0.3)	16 (9.9)	12 (8.1)	9 (5.9)	21 (7.0)
Musculoskeletal and connective tissue disorders	3 (1.8)	6 (3.6)	1 (0.6)	7 (2.1)	19 (11.8)	14 (9.5)	18 (11.8)	32 (10.6)
Back pain	0	0	0	0	9 (5.6)	3 (2.0)	7 (4.6)	10 (3.3)
General disorders and administration site conditions	1 (0.6)	22 (13.2)	27 (16.1)	49 (14.6)	11 (6.8)	25 (16.9)	16 (10.5)	41 (13.6)
Fatigue	0	9 (5.4)	10 (6.0)	19 (5.7)	2 (1.2)	5 (3.4)	4 (2.6)	9 (3.0)

Abbreviations: N=sample size of group; n=number of individuals occurring per adverse event category

The *severity of TEAEs* in the active total group was mild in 103 (30.7%), moderate in 158 (47.2%), severe in 60 (17.9%), and life-threatening in 2 (0.6%) participants. Two life-threatening TEAEs occurred in the 50 mg/day group: ischemic stroke (unlikely related), and hyponatraemia (possibly related). One TEAE in the 50 mg/day group was fatal (not related).

More patients had TEAEs of moderate and severe intensity in the 50 mg/day group (moderate: 49.4%, severe: 21.4%) as compared to the 30 mg/day group (moderate: 44.9%, severe: 14.4%), indicating a relationship between the dose and the intensity of TEAEs experienced.

The incidence of TEAEs by maximum intensity by treatment phase in study -004 indicates a higher proportion of placebo- and blarcamesine-treated participants with severe TEAEs in the maintenance phase (6.8% and 15.9%) compared to the titration phase (0.6% and 5.1%), likely a consequence of the longer treatment exposure in the maintenance phase.

No severe TEAE (by preferred term) was reported by more than 5% of blarcamesine-treated participants. Severe dizziness was experienced by 2.4% of blarcamesine-treated participants. Confusional state was severe in 0.6% of active total group patients.

Study drug related adverse events: 85.3% of TEAEs in the active total group were considered possibly, probably, or definitely related to blarcamesine. TEAEs from the nervous system disorders SOC, psychiatric disorders SOC, and General disorders and administration site conditions SOC were by majority rated at least possibly related to blarcamesine. TEAEs (by PT) most often considered as being at least possibly related in participants receiving blarcamesine were dizziness (44.8%), confusional disorder (21.8%), and balance disorder (11.4%).

When separated by titration and maintenance phase, a lower proportion of participants in the maintenance phase had at least 1 related TEAE than did participants in the titration phase in the blarcamesine 30 mg group (52.0% vs. 66.5%, respectively). In the blarcamesine 50 mg group, the proportions of participants who had at least 1 related TEAE in the maintenance phase and in the titration phase were similar (70.6% and 70.2%).

Study ANAVEX2-73-AD-EP-004

Common adverse events: The incidences of dizziness, confusional state, and fatigue were lower during the maintenance phase compared to the titration phase for both treatment groups. The incidences of COVID-19, urinary tract infection, and fall were higher in the maintenance phase than in the titration phase for both groups (Table 30).

Table 30. Study ANAVEX2-73-AD-EP-004: TEAEs in ≥5% of participants overall, by treatment group and study phase, OLE safety population

System Organ Class Preferred Term	Titration Phase			Maintenance Phase			Overall Total N = 300 n (%)
	Placebo to Blarcamesine N = 121 n (%)	Continued Blarcamesine N = 179 n (%)	Total N = 300 n (%)	Placebo to Blarcamesine N = 103 n (%)	Continued Blarcamesine N = 169 n (%)	Total N = 272 n (%)	
Subjects with at least one TEAE	**88 (72.7)**	**117 (65.4)**	**205 (68.3)**	**82 (79.6)**	**141 (83.4)**	**223 (82.0)**	**285 (95)**
Participants with TEAEs with frequency ≥5%	73 (60.3)	85 (47.5)	158 (52.7)	58 (56.3)	92 (54.4)	150 (55.1)	230 (76.7)
Psychiatric disorders	38 (31.4)	36 (20.1)	74 (24.7)	28 (27.2)	37 (21.9)	65 (23.9)	122 (40.7)
Confusional state	23 (19.0)	21 (11.7)	44 (14.7)	8 (7.8)	14 (8.3)	22 (8.1)	60 (20.7)
Anxiety	8 (6.6)	9 (5.0)	17 (5.7)	8 (7.8)	14 (8.3)	22 (8.1)	38 (12.7)
Agitation	1 (0.8)	4 (2.2)	5 (1.7)	8 (7.8)	5 (3.0)	13 (4.8)	18 (6.0)
Delirium	4 (3.3)	1 (0.6)	5 (1.7)	6 (5.8)	6 (3.6)	12 (4.4)	17 (5.7)
Nightmare	4 (3.3)	4 (2.2)	8 (2.7)	5 (4.9)	4 (2.4)	9 (3.3)	17 (5.7)
Depressed mood	6 (5.0)	3 (1.7)	9 (3.0)	2 (1.9)	4 (2.4)	6 (2.2)	15 (5.0)
Nervous system disorders	46 (38.0)	50 (27.9)	96 (32.0)	18 (17.5)	35 (20.7)	53 (19.5)	134 (44.7)
Dizziness	35 (28.9)	33 (18.4)	68 (22.7)	7 (6.8)	19 (11.2)	26 (9.6)	88 (29.3)
Headache	5 (4.1)	4 (2.2)	9 (3.0)	5 (4.9)	10 (5.9)	15 (5.5)	23 (7.7)
Somnolence	7 (5.8)	6 (3.4)	13 (4.3)	5 (4.9)	4 (2.4)	9 (3.3)	21 (7.0)
Lethargy	6 (5.0)	7 (3.9)	13 (4.3)	1 (1.0)	4 (2.4)	5 (1.8)	18 (6.0)
Balance disorder	3 (2.5)	7 (3.9)	10 (3.3)	2 (1.9)	5 (3.0)	7 (2.6)	17 (5.7)
Injury, poisoning and procedural complications	3 (2.5)	5 (2.8)	8 (2.7)	9 (8.7)	28 (16.6)	37 (13.6)	43 (14.3)
Fall	3 (2.5)	5 (2.8)	8 (2.7)	9 (8.7)	28 (16.6)	37 (13.6)	43 (14.3)
Infections and infestations	5 (4.1)	5 (2.8)	10 (3.3)	25 (24.3)	43 (25.4)	68 (25.0)	74 (24.7)
Urinary tract infection	4 (3.3)	2 (1.1)	6 (2.0)	14 (13.6)	23 (13.6)	37 (13.6)	40 (13.3)
COVID-19	1 (0.8)	3 (1.7)	4 (1.3)	11 (10.7)	25 (14.8)	36 (13.2)	39 (13.0)
Gastrointestinal disorders	14 (11.6)	8 (4.5)	22 (7.3)	9 (8.7)	16 (9.5)	25 (9.2)	43 (14.3)
Constipation	4 (3.3)	5 (2.8)	9 (3.0)	8 (7.8)	13 (7.7)	21 (7.7)	29 (9.7)
Nausea	11 (9.1)	3 (1.7)	14 (4.7)	2 (1.9)	4 (2.4)	6 (2.2)	19 (6.3)
General disorders and administration site conditions	8 (6.6)	11 (6.1)	19 (6.3)	3 (2.9)	5 (3.0)	8 (2.9)	24 (8.0)
Fatigue	8 (6.6)	11 (6.1)	19 (6.3)	3 (2.9)	5 (3.0)	8 (2.9)	24 (8.0)

OLE = open-label extension; TEAE = treatment-emergent adverse event.
Source: ANAVEX2-73-AD-EP-004

Of the 95% of participants who had at least one TEAE, 31.3% had TEAEs of mild and 42% had TEAEs of moderate *severity*. Severe TEAEs were reported by 21% of participants overall, and the incidence of was similar between the treatment groups in both phases of the study. Severe TEAEs that occurred in more than 2 participants were:

delirium (6 participants), agitation (3), urinary tract infection (3), orthostatic hypotension (4), neuropsychiatric symptoms (6), aggression (4), fall (5), orthostatic hypotension (4), syncope (3), femoral neck fracture (3), and urinary tract infection (3).

For most SOCs (except eye disorders), the incidence of severe TEAEs was higher in the maintenance phase than in the titration phase. Two participants (0.7%) had a life-threatening TEAE (small intestinal perforation and gastrointestinal carcinoma), and one participant (0.3%) had a fatal TEAE (fall). None of these events was considered as related to study drug. Another participant died during the OLE due to posterior cortical atrophy that was reported as a TEAE in the parent study and worsened during the

OLE; the TEAE is attributed to the parent study.

TEAEs *related to study drug* in ≥5% of participants overall were dizziness (25.7%), confusional state (16.7%), anxiety (6.3%), fatigue (6.0%), and somnolence (5.3%).

The incidences of drug-related dizziness, confusional state, and fatigue were higher in the titration phase relative to the maintenance phase and were higher in the placebo to blarcamesine group relative to the continued blarcamesine group during the titration phase.

Studies ANAVEX2-73-AD-004 and ANAVEX2-73-AD-EP-004 – side by side

The most frequently reported TEAEs in the double-blind phase were dizziness and confusional state and the incidence of these events was higher as compared to the extension study EP-004. The same observation was made for the remaining PTs under the nervous system disorders SOC, as well as for the PTs abnormal dreams, depressed mood, disorientation, fatigue, feeling abnormal, and nausea.

TEAEs with incidences >10% and reported more frequently in the OLE study were fall (14.3%), anxiety, COVID-19, and urinary tract infection (13.0% each). The incidence of these events in blarcamesine-treated participants of the double-blind study were 6.6%, 9.9%, 1.5%, and 5.1%, respectively. The high incidence of COVID-19 infections in the OLE study vs. the DB phase of study -004 relates to the timing of study EP-004 that coincided with the beginning of the pandemic in March 2020.

Except for the TEAEs noted above (dizziness, balance disorder, COVID-19, urinary tract infection, and fall), the incidences of TEAEs in the 2 studies were similar in blarcamesine-treated participants, and generally lower in placebo-treated participants.

Table 31. Studies -004 and EP-004: TEAEs in ≥5% of participants in any group, safety population

	Double-Blind Safety Population		Extension
System Organ Class Preferred Term	**ANAVEX2-73-AD-004 Placebo N=168 n (%)**	**ANAVEX2-73-AD-004 Blarcamesine Total N=335 n (%)**	**ANAVEX2-73-AD-EP-004 Blarcamesine Total N=300 n (%)**
Participants with at least one TEAE	129 (76.8)	324 (96.7)	286 (95.3)
Nervous system disorders	41 (24.4)	241 (71.9)	167 (55.7)
Dizziness	15 (8.9)	155 (46.3)	88 (29.3)
Balance disorder	3 (1.8)	39 (11.6)	17 (5.7)
Headache	8 (4.8)	30 (9.0)	23 (7.7)
Lethargy	3 (1.8)	26 (7.8)	18 (6.0)
Somnolence	5 (3.0)	26 (7.8)	21 (7.0)
Psychiatric disorders	43 (25.6)	201 (60.0)	179 (59.7)
Confusional state	5 (3.0)	80 (23.9)	58 (19.3)
Anxiety	6 (3.6)	33 (9.9)	39 (13.0)
Agitation	2 (1.2)	15 (4.5)	17 (5.7)
Delirium	6 (3.6)	7 (2.1)	16 (5.3)
Nightmare	3 (1.8)	16 (4.8)	16 (5.3)
Abnormal dreams	0	25 (7.5)	14 (4.7)
Depressed mood	4 (2.4)	21 (6.3)	15 (5.0)
Disorientation	0	21 (6.3)	14 (4.7)
General disorders and administration site conditions	12 (7.1)	83 (24.8)	59 (19.7)
Fatigue	2 (1.2)	28 (8.4)	24 (8.0)
Feeling abnormal	0	17 (5.1)	11 (3.7)
Gastrointestinal disorders	41 (24.4)	82 (24.5)	78 (26.0)

	Double-Blind Safety Population		Extension
System Organ Class Preferred Term	**ANAVEX2-73-AD-004 Placebo N=168 n (%)**	**ANAVEX2-73-AD-004 Blarcamesine Total N=335 n (%)**	**ANAVEX2-73-AD-EP-004 Blarcamesine Total N=300 n (%)**
Nausea	13 (7.7)	31 (9.3)	18 (6.0)
Constipation	5 (3.0)	19 (5.7)	25 (8.3)
Infections and infestations	26 (15.5)	75 (22.4)	111 (37.0)
COVID-19	4 (2.4)	5 (1.5)	39 (13.0)
Urinary tract infection	7 (4.2)	17 (5.1)	39 (13.0)
Injury, poisoning and procedural complications	22 (13.1)	32 (9.6)	67 (22.3)
Fall	16 (9.5)	22 (6.6)	43 (14.3)

TEAE = treatment-emergent adverse event.
Note: Active drug is ANAVEX2-73 (various doses).

Study ANAVEX2-73-003

The *most common TEAEs* were dizziness (37.5%), fall and constipation (29.2% each), upper respiratory tract infection (25.0%), UTI, headache, anxiety, rash (20.8% each). Confusional state and nausea occurred in 16.7% of patients each.

The majority of TEAEs was assessed as either mild or moderate in *severity*, and there was no clear association between dose and maximum intensity of TEAEs. Overall, 17 TEAEs in 8 participants were Grade ≥3 (one TEAE was Grade 5 [fatal TEAE of dementia]; the remaining were Grade 3).

The majority of dizziness TEAEs were Grade 1. One TEAE of dizziness was Grade 3 (severe), occurred in a participant between Day 1 and 15 of treatment, and was assessed as probably related to study drug. Confusional state was mainly mild and moderate in severity, and severe confusional state was reported in one patient on Day 3 (possibly related). One participant experienced four TEAEs of fall, and another experienced two TEAEs of fall. The majority of fall TEAEs was Grade 1, and all were assessed as not related to study drug. All TEAEs of constipation were Grade 1 and assessed as not related to study drug. One participant had four TEAEs of upper respiratory infections, and another had three TEAEs of upper respiratory infections. The majority of upper respiratory tract infections were Grade 1, and all were assessed as not related to study drug.

The only severe event that occurred in >1 participant was pneumonia (n=2); however, both cases were assessed as not related to study drug and both participants recovered.

The most common *treatment-related TEAE* was dizziness (25%). Six treatment-related TEAEs of increased lipase were seen in 3 participants (blarcamesine 30 mg dose; 2 participants at the 10 mg dose); 2 events in 1 participant were Grade 3 in intensity resulting initially in drug interruption and subsequently withdrawal from the study.

Three treatment-related TEAEs of peripheral oedema were reported in 3 participants (blarcamesine 20 mg dose and 2 participants with 10 mg dose); all 3 events were Grade 1 in intensity.

Three treatment-related TEAEs of confusional state were reported in 2 participants (blarcamesine 30 mg dose and the 50 mg dose); 1 event was Grade 3 in intensity.

Study ANAVEX2-73-002

In Part A, the most *common TEAEs* were dizziness (43.8%), headache (31.2%), and lethargy (21.9%). All TEAEs were mild or moderate *in severity*. TEAEs *related to study drug* in Part A of the study were

reported in 81.2% of participants overall. The most common TEAE related to study drug administration in Part A was dizziness (43.8%), followed by headache (21.9%), lethargy (18.8%), and unsteady gait and light-headedness (15.6% each).

In Part B, the most *common TEAEs* were dizziness (30.0%), upper respiratory tract infection (13.3%), urinary tract infection, and headache (10.0% each). A majority of TEAEs were mild or moderate in *severity*. Five severe TEAEs were reported for three participants (one participant each with blarcamesine 20 mg, 30 mg, and 50 mg): one patient reported a severe TEAE of *confusion* at the 50 mg dose (possibly related), one patient reported three severe TEAEs at 20 mg (dizziness on Day 1, probably related; serum amylase increased and lipase increased, both probably related and reported on Day 178), and the third patient reported a severe TEAE of delirium at 30 mg (not related).

TEAEs *related to study drug* administration in Part B of the study were reported in 53.3% of participants overall. Considering all participants, the most common TEAE related to study drug administration in Part B was dizziness (23.3%), followed by lethargy and confusion (6.7% each).

Study ANAVEX2-73-001

The most frequently reported TEAE with blarcamesine was dizziness (11/41, 26.8%). Three of 8 participants experienced 2 episodes of dizziness. Eight TEAEs of dizziness were mild, and three were moderate in severity. Dizziness was found dose-related, 25% of participants with 30 mg/day to 100% of participants with 60 mg/day. Headache was the second most common TEAE (17.0%).

78.0% of TEAEs were mild, and 22.0% were moderate in severity. No severe TEAEs were reported.

2.6.8.3. Serious adverse event/deaths/other significant events

Deaths

Four deaths have been recorded during the clinical development program.

ANAVEX2-73-AD-004

Participant 115-001 (50 mg dose group); a 73-year-old White male participant experienced a urinary tract infection resulting in urosepsis leading to hospitalisation 288 days after the first blarcamesine dose. The patient was treated with several IV antibiotics. A few days later, the patient was discharged from hospital to hospice and died as a consequence of **urosepsis**. Study drug was withdrawn as the participant was too unwell and was unable to swallow. The event of urosepsis was not rated related to blarcamesine. Prior to this event, non-serious TEAEs of acute renal failure and acute urinary retention (unrelated to treatment) were reported.

ANAVEX2-73-AD-EP-004

Participant 101-017: a 64-year-old Caucasian male treated with placebo during study -004. After 9 months, he experienced a SAE of **worsening of posterior cortical atrophy**. Over the course of one month, the participant was admitted to the hospital, released to "hospital at home", readmitted to the hospital and then discharged to a rehabilitation hospital for sub-acute rehabilitation. Approx. 6 weeks later, the participant rolled over into the OLE and initiated treatment with blarcamesine 10 mg/day. The participant was discharged 3 days later from the hospital to home care with services with the event ongoing. About 3 months later, worsening of posterior cortical atrophy was reported with a fatal outcome. No action was taken with blarcamesine. Initially, the event was rated as possibly related to study drug but reassessed as not related during follow-up.

Participant 113077: a 73-year-old female previously participated in study -004 where she received blarcamesine 30 mg/day; treatment with blarcamesine in the OLE began on 27 Jul 2021. On 14 Sep 2022 (Day 415, OLE), the participant had a fatal SAE of a **fall** with C1/C2 spinal injury and died on the same day. The event of fall was unlikely related to the study drug.

ANAVEX2-73- 003

Participant 103004: received 30 mg/day blarcamesine; died during the study (not related) due to dementia. The participant had been hospitalised for SAEs of urinary tract infection (Days 1706 to 1719) and pneumonia (Days 1722 to 1737) (study days are cumulative from the start of study -002). The participant's condition deteriorated following these hospital admissions. Study drug was discontinued on Day 1753 and the participant died on Day 1757 with the presumed cause of death being **end-stage dementia** due to Alzheimer's disease.

Serious Adverse Events

ANAVEX2-73-AD-004

56 patients (16.7%) experienced 77 SAEs in the blarcamesine active total group while 17 patients (10.1%) experienced 20 SAEs in the placebo group. The incidences in the 2 blarcamesine dose groups were similar. All individual SAEs (by PT) were experienced by <5% of blarcamesine-treated participants. SAEs reported in more than 1 blarcamesine-treated participant were: dizziness (5 participants, 1.5%), hyponatraemia (4 participants, 1.2%), syncope (3 participants, 0.9%), and seizure, transient ischemic attack, inguinal hernia, UTI, urosepsis, and delirium (2 participants each, 0.6%). All SAEs (by PT) in the placebo group were reported in only 1 participant except for pneumonia (2 participants, 0.2%). 19 patients (5.7%) on blarcamesine versus 5 patients (3%) on placebo experienced an SAE from the Nervous system disorders SOC.

Dizziness was the most frequent SAE with blarcamesine, all of which were reported recovered/ resolved; 1 was definitely related to study drug; 3 were possibly or probably related to study drug; and 1 was unlikely related. Study drug was interrupted in 4 patients and withdrawn for 1. The SAEs of dizziness were transient in duration (ranging from 3 days to 13 days).

When separated by treatment phase, 3.3% and 15.6% of patients from the blarcamesine active total group reported at least 1 SAE in the titration and maintenance period.

ANAVEX2-73-AD-EP-004

SAEs were reported for 25.7% of participants and the incidence was similar between treatment groups. The most common SAEs were delirium (7 participants, 2.3%), neuropsychiatric symptoms (6 participants, 2.0%), urinary tract infection (4 participants, 1.3%), and aggression and femoral neck fracture (3 participants each, 1.0%). When separated by treatment phase, 2.7% and 25.7% of patients reported at least 1 SAE in the titration and maintenance period, i.e. a majority of SAEs in the OLE occurred in the maintenance period.

In 9 of 77 participants with SAEs, the event was considered to be related to study drug, 6 from the placebo to blarcamesine group (depression, delirium, cerebrovascular accident, UTI, somnolence, and seizure) and 3 from the continued blarcamesine group (constipation, delirium, aggression).

ANAVEX2-73-003

58.3% (14 patients) experienced 30 SAEs (including bladder adenocarcinoma stage unspecified, neuropsychiatric symptoms, bradycardia, post procedural hypotension, nephrolithiasis, influenza, myelopathy, UTI (2), fall (2), skin abrasion, haematoma, pneumonia (2), dementia (2), Hodgkin's

disease, pelvic fracture, abdominal pain, uterine leiomyoma, confusional state, psychotic disorder, vaginal haemorrhage, agitation, panic attack, cholecystitis acute, ischemic strike, ankle fracture, and hypertension). None was rated as related to study drug. Three SAEs in two participants had an onset during study -002.

ANAVEX2-73-002

SAEs were reported for 1 and 4 participant(s) in Part A and Part B. An SAE of delirium was reported in 2 participants, each of whom had study drug discontinued due to the event. One of these events was moderate (possibly related to study drug), and 1 was severe in intensity (not related to study drug). The remaining SAEs were Hodgkin's lymphoma, UTI, and ankle fracture (all in Part B).

2.6.8.4. Laboratory findings

Mean changes over time (haematology, chemistry, and urinalysis)

Study ANAVEX2-73-AD- 004 and Study ANAVEX2-73-AD-EP-004

During the pivotal study -004 and its extension EP-004, no clinically significant within-group changes from (OLE) baseline or between-group differences in change from (OLE) baseline were observed over time in mean haematology, chemistry, or urinalysis laboratory values. For gamma glutamyl transferase (GGT), there was an increase at Week 48 in the blarcamesine 50 mg/day group only (mean change from baseline was 19.2 U/L).

Study ANAVEX2-73-003, Study ANAVEX2-73-002, and Study ANAVEX2-73-001

No clinically significant changes from baseline values were observed over time in mean haematology or chemistry laboratory values in these studies. Urinalysis results were not clinically significant (not collected in study -003).

Summary of Qualitative Urinalysis Laboratory Data

No notable differences from baseline in qualitative urinalysis parameters were observed for either treatment group in studies -004 and EP-004.

Shifts over time (haematology, chemistry, and urinalysis)

Study ANAVEX2-73-AD-004

Shifts in clinical haematology values from normal to low, normal to high, high to normal, or low to normal in >5% of participants in the placebo, blarcamesine 30 mg, and blarcamesine 50 mg groups mainly pertained to decreases in erythrocyte counts and concomitant decreases in haemoglobin and haematocrit, with possible compensation by increases in MCV.

There were no shifts in chemistry values (from normal to low, normal to high, high to normal, or low to normal) in >5% of participants in the placebo group, while shifts in the blarcamesine 30 mg and blarcamesine 50 mg groups (mainly for bicarbonate, creatinine, GGT, phosphate, and sodium) did not follow a specific pattern.

Study ANAVEX2-73-AD-EP-004

Shifts in clinical haematology values from normal to low or normal to high in >5% of participants in either treatment group mainly pertained to decreases in erythrocyte counts and concomitant decreases in haemoglobin and haematocrit, with possible compensation by increases in MCV.

Shifts in clinical chemistry values from normal to high were noted for ALT, AST, alkaline phosphatase creatinine, GGT, phosphate, and potassium (in less than 10% of patients in either treatment group);

however, mainly after Week 96 visits, when the number of patients substantially decreased. Shifts from normal to low in bicarbonate values were observed in participants in both treatment groups at all time points up to Week 96.

Study ANAVEX2-73-003, Study ANAVEX2-73-002, and Study ANAVEX2-73-001

Clinically significant shifts for haematology or chemistry laboratory values were either absent (study -003) or have not been analysed (studies -002 and -001).

Individual clinically significant abnormalities

The incidence of haematology, chemistry, or urinalysis laboratory abnormalities reported as TEAEs was low in study ANAVEX2-73-AD-004 (all were 0.3% in participants receiving blarcamesine) and ranged from 0.3% to 2.0% of participants overall in study ANAVEX2-73-AD-EP-004. ALT and/or AST increased as TEAE was reported in 2% of all blarcamesine treated patients in study EP-004. No participant had values meeting potential Hy's law criteria.

In study ANAVEX2-73-003, no TEAE was reported in more than 1 participant overall, except for lipase increased and hypercholesterolaemia (12.5% and 8.3% of patients). No apparent dose relationship was observed. There were 9 patients with 21 TEAEs that were associated with laboratory values (blood bicarbonate increased, blood potassium increased, lipase increased [6], blood creatinine increased, blood urea increased, GGT increased, hypercholesterolaemia [2], blood cholesterol increased, amylase increased [4], hyperglycaemia, hypoglycaemia, prostatic specific antigen increased).

In study ANAVEX2-73-002, TEAEs associated with laboratory values were reported for 4 participants: Two participants had events of hypercholesterolemia, including 1 participant with a concurrent event of hyperglycaemia. One patient was reported with two TEAEs on several occasions (serum amylase increased and lipase increased), and another patient had serum potassium increased and serum bicarbonate increased.

Vital signs, physical findings, and other observations related to safety

Mean changes over time

According to the applicant, no clinically significant trends over time in vital signs were observed and no notable differences were observed between placebo and the two blarcamesine groups in study ANAVEX2-73-AD-004. In study ANAVEX2-73-AD-EP-004, mean values for vital sign parameters at each post-OLE Baseline time point were similar to the OLE baseline values and no notable differences were observed between the placebo to blarcamesine and the continued blarcamesine groups.

Changes in electrocardiogram (ECG) parameters from baseline to Week 24/ Week 48 in study -004 did not follow a clear pattern, and mean values at each post-OLE Baseline time point in study EP-004 were similar to the OLE baseline values and no notable differences were observed between the placebo to blarcamesine and the continued blarcamesine groups.

Study ANAVEX2-73-002 and Study ANAVEX2-73-003

Statistically significant increases from baseline in respiratory rate and statistically significant decreases in systolic blood pressure were observed during Part A and Part B in study -002, but changes were not clinically significant. In Study -003, no clinically relevant changes in vital signs were observed according to the applicant.

Qualitative changes over time

In study ANAVEX2-73-AD-004, there were no clinically significant trends over time in physical or neurological examination findings. Physical and neurological examination at each post-OLE baseline time point in study ANAVEX2-73-AD-EP-004 did not worsen significantly compared to OLE baseline

showing a similar incidence of clinically abnormal findings for the two treatment groups.

Abnormal clinically significant ECGs were not reported at baseline in study -004 and occurred at Week 24 and Week 48 in a single subject each in the blarcamesine groups and in no patient from the placebo group. The proportion of participants with a clinically significant ECG overall interpretation at each post-OLE baseline time point was similar to that at the OLE baseline for each treatment group in EP-004.

No participants in either treatment group in study -004 and its EP-004 had a C-SSRS score of 4 or 5 at any post-baseline time point for either suicidal ideation or suicidal behaviour.

Study ANAVEX2-73-003

No clinically meaningful trends in ECG findings were observed. Two participants (treated with blarcamesine 10 mg and 20 mg) with normal ECGs at baseline had an overall ECG assessment reported as clinically significant.

Shifts over time

In Study ANAVEX2-73-AD-004, no clinically significant shifts over time in physical and neurological examination results or in ECG findings were observed from baseline to Week 48. A QTcF categorical analysis revealed a similar (low) number of patients in each treatment group with any post-baseline QTcF value >500 msec. A change from baseline of >30 - <60 msec and ≥60 msec to any time post-baseline was found slightly higher for the blarcamesine total active group as compared to placebo.

In Study ANAVEX2-73-AD-EP-004, shifts in physical and neurological examination findings as well as in ECG interpretation from normal (abnormal, not clinically significant) at baseline to clinically significant at a post-OLE baseline time point were low and similar in both treatment groups. Irrespective of ECG ratings at OLE baseline, more than 50% of patients in either treatment group had abnormal NCS ECGs at any post-baseline visit. 3.3% of patients in the continued blarcamesine group had a post OLE baseline QTcF of >500 msec vs. 1.2% of patients in the placebo to blarcamesine group. 4.6% of patients had a post OLE baseline increase of ≥60 msec in QTcF in the continued blarcamesine group vs. no patient in the placebo to blarcamesine group.

Study ANAVEX2-73-003

All 24 participants had QTcF values ≤450 msec at baseline. At any time post baseline, half of them remained on a QTcF ≤450 msec, while 8 participants (33.3%) had a QTcF values >450 msec and <480 msec, and 4 participants (16.7%) had a QTcF value >500 msec. Changes of ≥60 msec from baseline to any time post-baseline were recorded for 20.8% of the overall population.

Individual clinically significant abnormalities

The incidence of vital sign, physical finding, ECG, and other abnormalities reported as TEAEs within the investigations SOC was low (all were 0.3% in participants receiving blarcamesine) and not different to patients receiving placebo in study ANAVEX2-73-AD-004 and ranged from 0.3% to 2.7% of participants overall in study ANAVEX2-73-AD-EP-004.

Study ANAVEX2-73-AD-003, Study ANAVEX2-73-AD-002, and Study ANAVEX2-73-AD-001

The incidence of vital sign, physical finding, ECG, and other abnormalities reported as TEAEs was low without any clear pattern.

2.6.8.5. Safety in special populations

No information was provided by the applicant concerning safety in special populations.

No **pregnancy** was reported in patients treated with blarcamesine in the clinical programme.

No information is available concerning the effects of **overdose** of blarcamesine, nor are there any known antidotes to overdose with blarcamesine.

Based on its mechanism of action, the applicant assumes that blarcamesine is not expected to have **drug abuse** potential and has not been demonstrated to have any effects upon **withdrawal** of the drug, or any **rebound** effects.

Blarcamesine might has a temporarily minor to moderate influence on the **ability to drive and use machines**. Dizziness, confusion, and fatigue were reported in clinical trials, particularly during the initiation of treatment. Patients should be advised to exercise caution when driving or operating machinery, particularly during the first weeks of treatment or after a dose increase.

2.6.8.6. Safety related to drug-drug interactions and other interactions

No *in-vivo* drug-drug interaction studies were performed in humans. Drug-drug interactions were neither collected as part of the pivotal study -004 nor its extension study EP-004.

A number of *in-vitro* drug-drug interaction studies were conducted to evaluate the potential for blarcamesine and its active metabolite ANAVEX19-144 for potential DDI.

Study ANAVEX2-73 M12: CYP phenotyping to evaluate biotransformation of blarcamesine, specifically in the formation of the N-desmethyl metabolite ANAVEX19-144; as a result, studies in human liver microsomes showed that blarcamesine is transformed to its metabolite ANAVEX19-144 by CYP 3A4/5 (35.3%), 2C19 (31.6%), and 2B6 (19.3%).

The potential of blarcamesine and ANAVEX19-144 to reversibly *inhibit* CYP 1A2, 2B6, 2C8, 2C9, 2C19, 2D6 and 3A4 was evaluated in Study ANAVEX2-73 M04. Based on these results, blarcamesine and ANAVEX19-144 are not expected to show any clinically meaningful relevant DDI.

Moreover, based on the assessment of the potential of blarcamesine to cause intestinal CYP3A inhibition, the applicant concluded that blarcamesine is not expected to cause a clinically meaningful DDI with intestinal CYP3A.

The potential for blarcamesine (but not for ANAVEX19-144) to *induce* CYP1A2, CYP2B6, and CYP3A4 enzyme activity was determined in-vitro in Study ANAVEX2-73M03 (and ANAVEX2-73 M05). Activity measurements of CYP1A2, CYP2B6 and CYP3A4 following 48 hours of incubation with up to 5 µM of blarcamesine did not provide any indication of a clinically relevant CYP enzyme induction potential of blarcamesine.

The potential of blarcamesine to act as a substrate for the gastrointestinal efflux transporters (P-gp and BCRP) was also studied *in-vitro*: Blarcamesine was highly permeable in Caco-2 cell monolayer and was not a substrate for P-gp or BCRP in vitro. No other studies were conducted to assess the potential of blarcamesine or ANAVEX19-144 to cause interactions with hepatic or renal uptake transporters.

2.6.8.7. Discontinuation due to adverse events

Study ANAVEX2-73-AD- 004

108 (32.2%) participants treated with blarcamesine vs. 12 (7.1%) participants receiving placebo had at least 1 TEAE leading to treatment discontinuation, and the incidence was higher for the 50 mg/day group as compared to 30 mg/day (39.9% vs. 24.6%). Dizziness (8.1%) and confusional state (4.2%) were the most frequent TEAEs leading to treatment discontinuation in participants treated with

blarcamesine, followed by balance disorder (2.7%), anxiety (2.4%), and fatigue (2.1%).

TEAEs leading to treatment discontinuation were less frequently reported in the titration phase (56 participants; 16.7%) as compared to the maintenance phase (64 participants; 21.3%).

Despite the high incidence of TEAEs leading to treatment discontinuation, there were also frequent drug modifications in line with dose changes and drug interruptions due to TEAEs (45.7% and 31.3% of patients treated with blarcamesine and 7.1% and 9.5% of patients treated with placebo). These drug modifications were mainly related to the Nervous System Disorders SOC (dizziness, balance disorder, somnolence) and the Psychiatric Disorders SOC (confusional state).

A similar number of patients in the active blarcamesine group had TEAEs leading to drug modification during the titration and the maintenance phase (57% and 55.5%).

Study ANAVEX2-73-AD-EP-004

74 of 300 patients (24.7%) in this OLE discontinued due to a TEAE. Overall, a higher proportion of participants in the placebo to blarcamesine group had TEAEs that led to study drug discontinuation compared with the continued blarcamesine group (31.4% vs. 20.1%). These were mainly reported from the Psychiatric Disorders SOC and the Nervous System Disorders SOC. There were more patients with a TEAE leading to study drug discontinuation during the maintenance phase as compared to the titration phase (16.9% vs. 9.7%)

Study ANAVEX2-73- 002 and ANAVEX2-73-003

Five participants prematurely discontinued study drug during study -002 due to TEAEs at blarcamesine doses between 10 and 50 mg/day (two patients with delirium, and presyncope, dizziness, lethargy, disorientation, and confusion in one patient each).

Three participants (12.5%) reported 4 TEAEs that led to discontinuation from the extension study -003: neuropsychiatric symptoms (worsening behavioural and psychiatric symptoms of dementia; not related); TEAEs of amylase increased and lipase increased (possibly related); and ischemic stroke (not related).

2.6.9. Discussion on clinical safety

Safety database and exposure:

The main body of evidence with regards to the clinical safety of blarcamesine derives from the pivotal Phase 2b/3 study ANAVEX2-73-AD-004 with 48-week duration, which includes 508 patients with Early Alzheimer's disease. Of them, 335 patients received blarcamesine at the two target doses 30 mg/day and 50 mg/day and 168 received placebo and were included in the safety population.

Few supportive and uncontrolled short-term safety data (N=32 patients; enrolled in Part A of the Phase 2a study ANAVEX2-73-002) and long-term safety data (N=30 patients enrolled in Part B of ANAVEX2-73-002 and from those N=24 patients enrolled in the extension study ANAVEX2-73-003; doses of 10 to 50 mg/day) have additionally been provided.

The long-term safety of blarcamesine is mainly based on the open-label experience from study -EP-004, which offered 300 patients from study -004 up to 96 weeks of treatment (up to 144 weeks for patients in Australia) with blarcamesine at doses up to 50 mg/day or the best tolerated dose (121 patients, who were previously treated with placebo and 179 patients, who previously received blarcamesine). Upon request it has been clarified that all of these patients entered the extension period with a mean gap in dosing of ~16 days. Long-term safety is considered to be insufficiently presented in the dossier since patients with continuous blarcamesine treatment in the parent study -

004 and study EP-004 have not been analysed. The lack of longitudinal presentation could probably miss rare AEs that might be associated with longer duration of exposure to blarcamesine. Upon further request to evaluate these data, the applicant provided summary data of this group of patients. However, no source data have been presented with this summary, and the applicant did not explain the compilation of the summary. Moreover, the data presented appear highly conflicting with the data presented for the single studies -004 and EP-004. Since long-term safety is crucial for the benefit-risk of blarcamesine, this is a factor considered in the benefit-risk conclusions.

Moreover, 74 patients from studies -003 and EP-004 are currently included in a compassionate use programme (CUP) in Australia for which long-term safety data have been generated beyond those in the feeder studies (some patients received blarcamesine over 9 years). Upon request, the applicant clarified that only SAEs were to be collected in the CUP. However, no SAEs have been reported so far.

Both studies, -004 and EP-004, applied titration of blarcamesine up to 50 mg/day (or the best tolerated dose in study EP-004) followed by a maintenance period. Dose reductions due to safety/ tolerability issues were possible during the maintenance period.

In study -004, the initial titration was 2 or 3 weeks depending on the protocol versions. The 3-week up-titration accounted for emerging tolerability issues observed with the faster titration over 2 weeks. Upon request the applicant clarified that

- 7 patients have been enrolled under protocol version 1 (3 patient on placebo and 4 patients on blarcamesine active total), and 63 patients have been enrolled under protocol version 2 (21 patient on placebo and 42 patients on blarcamesine active total), both of which applied the 2-week titration schedule,
- 432 patients have been enrolled under protocol versions > 2 applying a 3-weeks titration (144 patient on placebo and 288 patients on blarcamesine active total).

The presentation of adverse events in the titration period of study -004 therefore mainly refers to patients with a 3-week titration schedule. Moreover, despite the lower numbers of patients included into the study under the protocol versions stipulating a 2-weeks titration, the 3-weeks titration period yielded a higher number of discontinuations due to adverse events (1/3 of patients in the blarcamesine group), which questions the benefit of a longer titration period to avoid CNS event-related discontinuations.

Moreover, a 4-week follow-up with dose reduction should have followed the double-blind period in study -004 for participants enrolled and treated in Australia under Protocol versions 1.0 and 2.0. Upon request, the applicant clarified that none of the patients consented under protocol versions 1.0 and 2.0 had a follow-up.

Study EP-004 applied a prolonged dose titration over 10 weeks for all participants, regardless of treatment assignment in study -004, due to the reporting of tolerability issues with a shorter up-titration of 2 to 3 weeks in study -004, i.e. starting at 10 mg/day for the first 2 weeks, then increasing the dose by 10 mg every 2 weeks over a 10-week period to a maximum maintenance dose of 50 mg/day or to the best tolerated dose (BTD). While it is understood that blarcamesine had to be titrated in patients, who previously received placebo during study -004, it remains unclear why patients treated with blarcamesine during the pivotal study obviously had to stop their treatment and were again re-titrated over 10 weeks in the OLE. Upon request, the applicant explained the re-titration was performed to maintain treatment assignment in the pivotal study concealed. This is not understood given that all patients in EP-004 received blarcamesine open-label. In addition, the applicant argued that patients assigned to placebo "at a higher dose" in -004 could not start dosing in EP-004 at the same blarcamesine dose, which is likewise not understood for an open-label designed study. The applicant was then asked to elaborate on the clinical consequences of completing the parent

study -004 with any doses higher than the starting dose of 10 mg/day in the extension study. With regard to safety, the applicant argued that the incidence in total TEAEs after switching from placebo to blarcamesine and with continuous blarcamesine treatment was similar during the titration period in the extension study. However, this explanation contrasts the overarching assumption that a prolonged titration period of 10 weeks in the EP-004 study at least reduces the prevailing CNS effects of blarcamesine.

The size of the safety database for blarcamesine comprises only 488 *individual* patients, who have been treated with blarcamesine across four clinical studies, including a negligible number of patients from study -002/ its long-term extension -003, while differences in the design (open-label) and patient population (patients with mild to moderate Alzheimer's disease) also need to be considered. The extent of the exposed patient population for the evaluation of clinical safety should be consistent with the recommendations in ICH E1. In this guideline, the minimum requirement for safety evaluation of an investigational drug is as follows: 100 patients treated for at least 1 year, 300-600 patients treated for at least 6 months, and 1500 individuals treated in total. The small safety database of blarcamesine is clearly insufficient with respect to the disease to-be treated, which is not rare, and - even more important – does not take into account the novel mechanism of action claimed by the applicant. Up till now, no other sigma receptor agonist has been approved, which additionally weakens the data package and strongly questions acceptability of one pivotal trial. Despite this, evaluable data were further reduced by the high number of study discontinuations in study -004 (twice as high in the total active group as compared to placebo; 40.2% vs. 20%), and higher with 50 mg compared to 30 mg. The main reason for discontinuations was adverse events, which questions the safe use of blarcamesine. Overall, this remains an unresolvable major concern, that is taken into account in the benefit-risk evaluation.

The majority of early discontinuations occurred during the maintenance phase (29.3% vs. 10.1% during the titration phase). Discontinuations during the maintenance period most frequently occurred up to Week 24. In addition, more than half of the 300 patients (57.3%), who entered the extension study EP-004, discontinued treatment at any time during the study; again, mainly due to TEAEs (more frequent in patients with placebo during the parent study) or withdrawal of consent (more frequent in patients continuously been treated with blarcamesine; primarily due to study burden or progression of AD that resulted in higher care for the patients). The high dropout rate in the OLE study despite the prolonged titration over 10 weeks questions whether this is a reasonable measure to improve tolerability.

The overall extent of exposure for blarcamesine based on the five clinical studies contributing to clinical safety has been provided upon request. Regarding the treatment exposure of the indicated dose range (blarcamesine 10-30 mg) proposed in the label, 263 patients and 206 patients had the treatment exposure duration of at least 6 months and 12 months, respectively, with 102 patients being exposed to the treatment for 24 months or longer.

In the pivotal study -004, the mean (SD) duration on study drug was lower in the combined dose groups as compared to placebo: 225.3 (145.49) days versus 295.6 (97.51) days.

The titration during the maintenance period in the pivotal trial has been insufficiently described. Despite prespecification of two target doses, i.e. 30 mg/day and 50 mg/day, a flexible dosing schedule has been applied, i.e., participants were allowed to down-titrate and up-titrate based on tolerability after the initial titration phase. Consequently, the average daily exposures in the 30 mg and 50 mg blarcamesine dose groups were comparable at each of the study visits, and the mean dose in each group was even less than 30 mg/day since a majority of patients had to adapt the dose due to tolerability reasons. Therefore, and even after the second CHMP request for clarification, the recommended maintenance dose of 30 mg/day in the label remains not appropriately justified.

Moreover, it remained unclear how the recommendation for the maximum daily dose of 40 mg in the label (not formally tested) is justified considering that the highest dose in the study was 50 mg/day. Meanwhile, the applicant abstained from pursuing the 50 mg dose and also the previously proposed 40 mg dose as maintenance/ maximum doses due to a lack of additional benefit, but worse tolerability as compared to the 30 mg dose. From a safety perspective, the applicant did not provide new comprehensive safety information for the patients in study -004, who reached and finally remained on the recommended 30 mg dose, neither for the overall AD population nor for the subpopulation of AD patients applied for.

The mean duration of exposure in the extension study EP-004 was 607±359 days in total, and substantially lower in blarcamesine-naïve patients as compared to patients with continuous treatment. A majority of patients in both groups (47.3%) was up-titrated to 50 mg/day during the 10 weeks titration phase and more than 50% of patients in either group remained on the final dose that has been reached during the titration phase. While this could be indicative of a better tolerability with prolonged titration, it needs to be put into perspective with the high discontinuation rate in the extension study.

Adverse events

The incidences of TEAEs, treatment-related TEAEs, SAEs, and TEAEs leading to treatment discontinuation with blarcamesine were clearly higher compared to placebo in study -004. A similar picture can be seen for the uncontrolled studies, while reporting of SAEs was higher during the long-term studies EP-004 and -003 as compared to the pivotal study -004.

Common adverse events

Adverse events in study -004 were mainly attributed to the Nervous System Disorders, Psychiatric Disorders, Gastrointestinal Disorders, General disorders and Administration Site Conditions, Infections and Infestations, Injury, Poisoning and Procedural Complications, and Musculoskeletal and Connective Tissue Disorders SOC.

The most common, specific, and dose-related TEAEs with (active total) blarcamesine in study -004 were in line with its CNS activity, i.e. dizziness (46.3% [39.5% and 53% in 30 mg and 50 mg groups, respectively]), confusional state (23.9% [25.6% and 22.2% in 30 mg and 50 mg groups, respectively]), and balance disorder (11.6% [9.6%, 13.7% in 30mg, 50 mg groups, respectively]). In the placebo group, the incidences of these events were 8.9%, 3%, and 1.8%, respectively. Their incidences in the blarcamesine groups were higher or similar during the titration phase as compared to the maintenance phase indicating that these events specifically occur during initiation of treatment. These events were mainly mild and moderate in severity, severe in few patients only (2.4%, 0.6%, and 0.3%), recorded as SAEs in 1.5%, 0%, and 0% of patients, and led to treatment discontinuation in 8.1%, 4.2%, and 2.7% of patients. In addition, nearly all TEAEs from the psychiatric disorders SOC were found dose-related (i.e. anxiety, abnormal dreams, depressed mood, disorientation, and nightmare). In addition, there was also a high rate of lethargy, i.e. 7.8% in the active treatment group compared to 1.8 % in placebo group. Moreover, the following events were more frequently reported in actively treated patients as compared to placebo: somnolence (7.8% vs. 3%), depressed mood (6.3% vs. 2.4%), fatigue (8.4% vs. 1.2%), and memory impairment 4.2% vs. 2.4%).

TEAEs related to CNS effects, i.e. nervous system disorders (e.g. dizziness, balance disorder) and psychiatric disorders (e.g., confusional state, anxiety), generally entailed frequent dose modifications, i.e. drug discontinuations (18.5% and 18.2% of patients in the active total blarcamesine group), dose changes (31.9% and 23.6%), and drug interruptions (20.3% and 11%), which was more pronounced with higher doses. Half of the patients with discontinuations due to CNS-related TEAEs did so during

the titration phase.

Other TEAEs occurred in <10% of participants treated with blarcamesine. The most common TEAEs in the placebo group were fall (9.5%) and dizziness (8.9%).

In study EP-004, common TEAEs were qualitatively the same as reported during study -004. The most common TEAEs (≥10% of participants overall) during the OLE study were dizziness (29.3%), confusional state (20.7%), fall (14.3%), urinary tract infection (13.3%), COVID-19 (13.0%), and anxiety (12.7%). The CNS-related events were also more frequently reported during the (prolonged) titration phase as compared to the maintenance phase, and more frequently in patients who switched from placebo to blarcamesine in this study compared to those with continuous treatment.

Based on the comparison of common TEAEs in study -004 (up to 48 weeks) and EP-004 (up to 96 weeks [up to 144 weeks for participants in Australia]), the safety profile does not appear to be much different during longer-term treatment. Higher incidences of TEAEs related to infections might be attributed to the longer treatment duration. However, the increased incidences of falls during longer treatment exposure, especially in the continued blarcamesine group, might be a consequence of the CNS effects of the drug worsening the underlying medical condition, which is of concern in this patient population.

TEAEs in study -004 were mainly mild and moderate in severity. Severe TEAEs were more frequent with the 50 mg dose as compared to the 30 mg dose (21.4% and 14.4%) and mainly derived from the Nervous System Disorders SOC (see above). Two life-threatening TEAEs occurred in the 50 mg/day group (ischemic stroke [unlikely related] and hyponatraemia [possibly related]. When separated by treatment phase, severe events occurred more frequently with longer exposure during the maintenance period. Longer exposure to blarcamesine in EP-004 did not relevantly increase the incidence of severe events. Two life-threatening TEAEs, i.e. small intestinal perforation and GI carcinoma, and one fatal TEAE (fall) were not rated as related to blarcamesine.

Study drug related adverse events/ Adverse drug reactions (ADRs) proposed in the SmPC

The majority of TEAEs with blarcamesine in study -004 was found treatment-related (85.4%), headed by TEAEs from the Nervous System Disorders SOC and Psychiatric Disorders SOC (dizziness, 44.8%; confusional disorder, 21.8%; and balance disorder, 11.4%). The incidence of treatment-related events was slightly lower during the OLE. During study -003, 3 patients experienced 6 TEAEs of increased lipase rated as related to blarcamesine. However, neither lipase nor amylase were collected routinely, which hampers a clear correlation with active study drug.

The following **ADRs** have now been proposed for blarcamesine: *dizziness, confusional state (each very common), somnolence, lethargy, balance disorder, headache, nausea, fatigue, anxiety, agitation, depressed mood, fall, UTI, and Upper respiratory tract infection (each common), and lymphopenia (uncommon, not agreed to be included)*. The methodology used to define ADRs proposed for inclusion in section 4.8 of the proposed SmPC has not been sufficiently described, although this had been specifically emphasised to the Applicant. In addition, some of the proposed ADRs (i.e. urinary tract infection, upper respiratory tract infection, and fall) were either not found to occur at a higher incidence in the blarcamesine group as compared to placebo or lack a clear dose-relation and have rarely been identified as TEAEs related to study drug. A comprehensive analysis of TEAEs, with >2% incidence in any of the treatment groups, and with a higher frequency in any of the blarcamesine dose groups than in the placebo group should have been conducted based on study -004. Results should then have been supplemented by additional information deriving from study EP-004 and by the results of any other clinical studies, if applicable.. Subsequently, all TEAEs should have been analysed for a causal relationship to blarcamesine based on temporal aspects and biologic plausibility.

A main shortcoming of the clinical safety evaluation is the lack of pre-specification and assessment of

Adverse Events of Special Interest (AESI), which would have been expected based on the proposed mechanism of action, nonclinical toxicology profile, and emerging clinical data.

Hence, the safety profile of blarcamesine has been insufficiently characterised especially with regard to its CNS activity, which remains of major concern:

As delineated in the *Guideline on the clinical investigation of medicines for the treatment of Alzheimer's disease* (CPMP/EWP/553/95 Rev.2), "*special efforts should be made to assess potential adverse effects that are characteristic of the class of drugs being investigated depending on the action on distinct receptor sites or enzymes (…)*". Moreover, the same guideline also stipulates that the effect of withdrawal should be systematically monitored.

The requirement to address the dependence potential for blarcamesine as a CNS active compound was highlighted during the pre-submission meeting since the applicant claims modulation of the Sigma 1 receptor (S1R) as a novel mechanism of action, thus lacking a licensed predecessor drug. While the dependence potential needs to be primarily investigated in preclinical studies, this has not been conducted by the applicant. In the nonclinical in vitro studies, blarcamesine and its active metabolite ANAVEX19-144 revealed high agonist affinity for S1R compared to S2R receptors at small micromolar concentrations and a 0.2- to 3.5-fold lower affinity at muscarinic M1 to M5 receptors. In addition, a similar high affinity could be shown for ANAVEX19-144 for the NMDA receptor and the μ-opioid receptor within the range of S1R and muscarinic cholinergic interactions, respectively. Therefore, it cannot be ruled out that the active metabolite contributes to a significant extent to the observed CNS effects in the nonclinical and clinical studies. Reference is made to the nonclinical assessment. Given that there is a broad scientific rationale for involvement of S1R agonism as well as μ-opioid receptor agonism in dependence and withdrawal (Aguinaga et al., 2019; Cobos et al., 2008; Sabino et al., 2017; Contet et al., 2004), specific monitoring of CNS-related events and outcomes would have been expected.

However, when applying definitions for events related to dependence (e.g. dizziness, somnolence, confusional state, disorientation, euphoric mood, aggression, hallucination, mood swings, mania, and feeling abnormal) and withdrawal effects (hypersomnia, anxiety, abnormal dreams/ nightmare, depression/ depressed mood, agitation, insomnia, sleep disorder, and restlessness) according to Cosci et al., 2020 and Sellers et al., 2018, these were more frequently reported for blarcamesine as compared to placebo. For most of the PTs potentially related to dependence and withdrawal, a dose effect can be observed, although, confirmation is hampered by various dose changes and interruptions during the study. Of important consideration, these side effects have an impact on the cognitive and behavioural burden of the underlying disease, which can hardly be acceptable for a drug claiming improvement of cognitive impairment.

Based on these considerations, additional analyses are still requested, while these data will further inform on adequate labelling in the product information and any additional risk minimisation measures. In this context, the applicant is first asked to present common adverse events for blarcamesine occurring in ≥2% of patients in any of the treatment groups for studies -004 and EP-004. Specifically, the analysis of the data for study -004 (the pivotal study) should serve as a basis for considering adverse events as ADRs in section 4.8 (see also section 4.10. of this AR). In a second step, the applicant is requested to thoroughly review CNS-related TEAEs indicative of dependence and withdrawal symptoms in study -004, supplemented by supportive data from the remaining clinical studies EP-004, -002, -003, and -001, as applicable, including time to onset, severity, discontinuation, interruption of treatment, dechallenge and rechallenge experience, duration etc. for the overall study population as well as for the SIGMAR1 wild-type genotype population, which is now claimed as the proposed target population. Consequently, the applicant should have provided adequate labelling in the clinical sections of the product information and proposed adequate risk minimisation measures to be

included in the RMP.

Serious adverse events and deaths, other significant events

A total of four **deaths** have been reported in the clinical programme, none of which was rated as related to blarcamesine by the investigator: urosepsis (50 mg/day blarcamesine group) was reported as a fatal event in study -004, and fall (resulting in C1/C2 spinal injury in a patient who received 30 mg/day blarcamesine) led to death in study EP-004. One patient (101-017) in study EP-004 died due to a SAE of worsening of posterior cortical atrophy that already emerged during the parent study when the patient was treated with placebo. Therefore, the assumption that this fatality is not related to blarcamesine cannot be firmly concluded. During study -003, (end-stage) dementia was the cause of death on study Day 1757 in a patient treated with blarcamesine 30 mg/day.

The incidence of **SAEs** was higher in the blarcamesine active total group as compared to placebo in study -004 (16.7% vs. 10.1%). The incidence of serious TEAEs in blarcamesine treatment groups was similar (15% in the 30 mg treatment group and 18.5% in the 50 mg treatment group). Reporting of SAEs was more frequent in the maintenance as compared to the titration period in line with the longer duration of exposure. 1/3 of patients with SAEs were reported with events from the Nervous System Disorders SOC, headed by dizziness (5 patients). All dizziness SAEs led to drug modification (either interruption or discontinuation), had a duration between 3 and 13 days, and all but one were rated as related to blarcamesine. There were also 3 patients reported with SAEs of syncope with blarcamesine, two of which had a preceding event of dizziness, and one SAE with loss of consciousness. These events might subsequently trigger further events, e.g. falls. Hyponatraemia was reported as SAE in 4 patients on blarcamesine (no SAE in the placebo group), basically during the first 6 weeks of treatment, more frequently with 50 mg/day as compared to 30 mg/day (3 vs. 1 patient) and possibly related to study drug in 2 patients. However, a mechanistic rationale could not be identified. The available data do not support a clear contribution of blarcamesine to these SAEs. Other SAEs (e.g. GI disorders, infections, cardiac disorders) were either reported with a similar incidence for placebo- and blarcamesine treated patients or in single patients only. In general, longer exposure to blarcamesine in EP-004 does not seem to increase the incidence of SAEs in general but, more likely as a consequence of worsening of the underlying disease and conditions related to an older patient population. 25.7% of participants were reported with SAEs during the OLE. Only 9 SAEs were assessed as related to blarcamesine in the OLE (depression, delirium [2], aggression, somnolence, constipation, cerebrovascular accident, UTI, and seizure). However, upon further review, a biological plausibility or mechanistic rationale appears unlikely for cerebrovascular accident, UTI, and seizure.

The high incidence of TEAEs leading to drug modification, especially to discontinuation of treatment, limits the validity of the safety database and questions the tolerability of blarcamesine: Overall, a high number of (dose-related) **discontinuations due to TEAEs** was recorded for blarcamesine in study -004 compared to placebo (32.2% vs. 7.1%), while the slightly higher rate of discontinuations due to TEAEs in the maintenance phase as compared to the titration phase is outweighed by the longer duration of exposure.

Moreover, dose changes and drug interruptions have been reported in 45.7% and 31.3% of the patients treated with blarcamesine in study -004. The main reason for these modifications was CNS-related TEAEs (dizziness and confusional state; see above). A slight overall reduction of discontinuations due to TEAEs was observed in the OLE, with a higher incidence in patients with placebo during study -004 as compared to those with continued blarcamesine. Evaluation and discussion of TEAEs leading to discontinuation with regard to severity, seriousness, follow-up/ outcome and discussion of narratives has been provided upon request for studies -004 and EP-004. Of the overall 182 patients out of 456 patients (40%), who discontinued treatment with blarcamesine during study -004 or EP-004, 55% were reported with psychiatric disorders, 46% with nervous system

disorders, and 13% with TEAEs from the General disorders and administration site conditions. Dizziness, mainly moderate in severity, led to discontinuation in 7.7% of all patients treated with blarcamesine in studies -004 and EP-004, and had an onset mainly during initiation or following dose increase. All events resolved following discontinuation. Dose reduction attempted in some of the patients prior to discontinuation was obviously insufficient. Confusional state led to discontinuation of blarcamesine in 5% of all patients in studies -004 and EP-004, was of moderate severity in all but one (severe), and resolved after discontinuation. The onset was not strictly related to treatment initiation; therefore, slower titration is not considered to mitigate discontinuations due to confusional state.

Balance disorder led to discontinuation of blarcamesine in 2.4% of all patients treated in studies -004 and EP-004, was often moderate in severity and resolved in all patients after discontinuation. Balance disorder events are thought to increase the risk for falls in this frail population. As a consequence, the applicant proposed an update of section 4.8 of the SmPC. However, the incidence of TEAEs leading to discontinuation in the description of selected TEAEs should only focus on the incidences in the pivotal trial.

Upon request, the applicant provided a tabular summary of TEAEs reported in study -004 by SOC, PT, and treatment group in order to evaluate the safety profile in **special populations** (ie, intrinsic factors, such as gender, ethnicity, race, and rs1800866 gene variant; extrinsic factors, e.g. region). However, even if analysis of TEAEs by age group (<75 vs. ≥75 years), sex and in genetic subpopulations vs. ITT did not reveal any clinically significant differences in the overall safety profile of blarcamesine, the presentation of data does not take into account the placebo group and is therefore still not sufficient. The same applies to the analysis by extrinsic factors that revealed an influence of comedications on the incidence of CNS-related TEAEs. Likewise, no studies were conducted in patients with renal or hepatic impairment, and the lack of this data has been therefore asked to be included as missing information in the RMP. The relevant information in SmPC sections 4.2, 4.3 (patients with severe renal or hepatic impairment are excluded from treatment with blarcamesine), 4.4, and 5.2 is not aligned and even contradictory, and it remains fully unclear whether there is any safety data regarding patients with mild, moderate or severe renal or hepatic impairment. Therefore, the adequacy of the information in the proposed SmPC cannot be judged at present.

The metabolism of blarcamesine has not been elucidated, except the initial demethylation resulting in the active metabolite ANAVEX19-144. Therefore, the evaluation of pharmacokinetic interactions was limited to inhibition of CYP isoenzymes by blarcamesine and its active metabolite while induction of CYP isoenzymes and interaction with gastrointestinal efflux transporters P-gp and BCRP was solely tested for blarcamesine. Thus, the potential for **drug-drug interactions** with blarcamesine and its active metabolite ANAVEX19-144 has been insufficiently characterised, which is especially concerning against the background of the intended patient population of advanced age likely receiving polypharmacy (**nonclinical/ clinical pharmacology MO**). As a consequence, the safety of blarcamesine related to drug-drug interactions has not been evaluated in the pivotal study.

Since CYP3A4 (and CYP2C19) were determined as the major enzymes contributing to the blarcamesine metabolism, the applicant should have provided a summary of clinical safety in patients in the clinical blarcamesine programme, who have concomitantly been treated at least with strong CYP3A4 inhibitors or CYP3A4 inducers. As a result, there were 2 cases of concomitant use of either a strong CYP3A4 inhibitor (erythromycin) or inducer (carbamazepine) with blarcamesine in Study ANAVEX2-73-AD-004 (ANAVEX2-73-AD-004-101003 and ANAVEX2-73-AD-004-101014). There were no SAEs reported in these subjects.

No clear safety signal derived from the provided haematology, chemistry evaluations, and urinalysis.

Mean *haematological* parameters were found similar for blarcamesine and placebo in study -004 (and in the two blarcamesine groups in study EP-004), while there were some shifts in the blarcamesine

group from normal to low and low to normal in erythrocytes, haematocrit and haemoglobin, as well as lymphocytes. Shifts from normal to low in lymphocyte counts were more frequently recorded in the continued blarcamesine group in the OLE as compared to the placebo-to-blarcamesine group and led to reporting of lymphocyte count decreased as TEAE in 5 patients. However, upon review, there seems to be no mechanistic rational for lymphocytopenia. Therefore, the applicant was asked to delete "lymphopenia" as ADR from SmPC section 4.8. Mean blood *chemistry* parameters were also similar for blarcamesine and placebo and shifts did not indicate a specific pattern. TEAEs associated with increases in serum amylase and lipase (not routinely measured in the blarcamesine clinical studies) were noted in studies -002 and -003 in a single patient rated as related to blarcamesine. However, a causal relationship could not be explained by the applicant. While the study protocols of studies -004 and EP-004 pre-specified coagulation parameters to be included in the clinical laboratory panel, no coagulation testing was performed and this has not been explained in the CSRs as protocol deviation, which is a clear shortcoming in the clinical documentation.

With regard to vital signs, discrete mean decreases in diastolic and systolic blood pressure and slight increases in heart rate up to Week 48 have been noted in the 50 mg/day group in study -004, supported by data in study EP-004 (changes appear slightly higher in the placebo to blarcamesine group as compared to the continued group) and in study -003. This finding, however, contrasts the observations in the nonclinical studies, where a dose-dependent, long-lasting (within 14 h post dose) and statistically significant increase in mean, systolic and diastolic arterial blood pressure after oral gavage doses ≥5 mg/kg blarcamesine was found in telemetered Beagle dogs (see nonclinical AR). Small shifts in systolic blood pressure from normal to high were similar for placebo and blarcamesine 30 mg and 50 mg dose groups in study -004. No significant shifts were reported for diastolic blood pressure in either treatment group. There were slightly more shifts in heart rate from normal to low in the blarcamesine dose groups as compared to placebo in study -004. No obvious trends in vital signs shifts with blarcamesine can be seen in study EP-004.

No significant **ECG changes** have been noted in the blarcamesine clinical studies. No significant mean changes from baseline to Week 24/ Week 48 in the QTcF interval were noted in either treatment group in study -004, which supports the absence of a clinically significant QTc prolongation at therapeutic doses in line with the preclinical results. Very few shifts (n=3) from normal or abnormal not clinically significant (NCS) at baseline to abnormal clinically significant post-baseline were noted in the blarcamesine groups with no worsening over time in study EP-004. Categorical analysis of QTcF at any time post-baseline was not significantly different for placebo and for the blarcamesine groups as were changes from baseline of >30 to <60 msec (16.6% vs. 13.5%) or >60 msec (4.1% vs. 3.2%). The relevance of a higher reporting of QTcF shifts during long-term extension study -003 remains uncertain but might be explained by concomitant medication (e.g. donepezil).

2.6.10. Conclusions on the clinical safety

Safety of blarcamesine in the target indication has not been sufficiently demonstrated, neither in the initially intended population nor in the restricted population applied for during the course of the procedure.

Overall, 488 individual patients with Early Alzheimer's disease and mild to moderate Alzheimer's disease were exposed to blarcamesine in the clinical programme across four clinical Phase 2/3 studies. The clinical safety evaluation, which informs the product information is mainly based on the 48-week data in the pivotal study ANAVEX2-73-AD-004, which includes 335 patients with at least one dose of blarcamesine at the target doses of 30 mg/day and 50 mg/day with a mean (SD) duration of exposure of 225.3 (145.49) days for both dose groups combined. However, there was a high amount of down-titration during the maintenance phase of the double-blind phase, which does not allow to conclude on

the adequacy of the recommended maintenance and also proposed maximum dose of 30 mg/day given that the average daily dose was below 30 mg at each visit in both blarcamesine dose groups.

Long-term safety from study ANAVEX2-73-AD-EP-004 adds between 96 and 144 weeks of treatment with blarcamesine in 179 patients with continued treatment from study -004, for whom safety has not been comprehensively summarised in a longitudinal analysis post-hoc. This applies to the overall AD population as well as for the SIGMAR1 wild-type genotype population, which is now claimed as the proposed target population.

Taking into consideration that dementia affects more than 55 million people worldwide with Alzheimer's disease accounting for up to 75% of the cases, the size of the safety database based on a single pivotal trial is clearly insufficient for marketing authorisation, especially considering the high dropout rate (40.2% in study -004 and 57% in study EP-004, mainly due to TEAEs) and the new mechanism of action claimed as sigma receptor agonist.

The main safety issue identified for blarcamesine is attributed to its CNS activity, i.e. to TEAEs from the Nervous System Disorders, Psychiatric Disorders, and General disorders and Administration Site Conditions SOC, specifically to TEAEs of dizziness (46.3%), confusional state (23.9%), balance disorder (11.6%), anxiety (9.9%), and fatigue (8.4%). While not uncommon in patients with dementia, these events are reported with higher incidences in the blarcamesine dose groups as compared to placebo and a correlation with the potential of blarcamesine and its pharmacologically active metabolite ANAVEX19-144 for dependence and withdrawal effects, cannot be excluded since it has been insufficiently characterised in the clinical blarcamesine programme. This applies to the overall AD population as well as for the SIGMAR1 wild-type genotype population, which is now claimed as the proposed target population. These events also frequently contributed to discontinuations from treatment, dose changes or drug interruptions in the clinical studies. Prolonged titration (over 10 weeks during the extension study EP-004) seems to mitigate but not to offset these effects given the even higher discontinuation rate during the OLE. Overall, the CNS effects can best be explained by the S1R agonism and the high affinity of ANAVEX19-144 to the NMDA and the μ-opioid receptor.

At present the ADRs identified by the applicant are *dizziness, confusional state (each very common), somnolence, lethargy, balance disorder, headache, nausea, fatigue, anxiety, agitation, depressed mood, fall, UTI, and upper respiratory tract infection (each common), and lymphopenia (uncommon)*. The primary source of evidence as well as the methodology used to define the ADRs for inclusion in section 4.8 of the SmPC has not been adequately described

2.7. Risk Management Plan

The CHMP, having considered the data submitted in the application was of the opinion that due to the concerns identified with this application, the risk management plan cannot be agreed at this stage.

2.8. Pharmacovigilance

2.8.1. Pharmacovigilance system

The CHMP considered that the pharmacovigilance system summary submitted by the applicant fulfils the requirements of Article 8(3) of Directive 2001/83/EC.

2.8.2. Periodic Safety Update Reports submission requirements

Not applicable.

2.9. Product information

2.9.1. User consultation

The Applicant has not submitted results of an appropriate user testing.

3. Benefit-Risk Balance

3.1. Therapeutic Context

3.1.1. Disease or condition

Dementia, or major neurocognitive disorder, is defined as a progressive loss of cognitive functioning which interferes with independence in daily activities, including occupational, domestic or social functioning.

It can be caused by a variety of disorders, including neurodegenerative diseases, and may be associated with other symptoms such as depression, anxiety, paranoia, delusions, visual hallucinations or apathy. Dementia occurs in about 1 to 2% of adults aged less than 65 years old but the prevalence increases with age and reaches 30 to 50% by age 85. As of 2019, dementia was affecting around 11 million people in Europe and 55 million people worldwide (Nichols et al., 2022; World Health Organization, 'Dementia: key facts' [Online]). With Europe's ageing population, the number of individuals affected by dementia is expected to reach 19 million by 2050 (Nichols et al., 2022). Dementia does not only affect patients but also has psychological, social and economic impacts for their relatives, their health carers.

Alzheimer's disease is the main cause of dementia affecting 60 to 80% of the individuals living with dementia (WHO, key facts; Alzheimer's disease facts and figures, '2018 Alzheimer's disease facts and figures', Mar. 2018, [Online]). AD is ultimately fatal.

In AD patients, biological changes start occurring years to decades before the onset of clinical signs and symptoms. During this so-called "pre-clinical phase", AD neuropathology is characterized by the aggregation of two proteins in the brain, β-Amyloid (Aβ) and Tau, leading to the deposition of extracellular Amyloid plaques and intracellular neurofibrillary tangles (NFTs), respectively. As the disease progresses, AD patients further display synaptic abnormalities, neuronal death, cognitive decline and memory impairment. At this clinical stage, two phases are distinguished. First, the prodromal AD, also referred to as mild cognitive impairment (MCI), is characterized by impaired cognitive abilities without loss of independent functioning. It is followed by "AD dementia", corresponding to the mild, moderate or severe loss of independent social and occupational functioning due to cognitive impairment.

Blarcamesine is a small molecule agonist of the Sigma 1 receptor (S1R) and, to a lower extent, a modulator of the muscarinic acetylcholine receptor (M1 to M5) that is proposed for treatment of AD and dementia in adults. Activation / modulation of S1R in the CNS by blarcamesine and its structurally related demethylated active metabolite ANAVEX19-144 are proposed to promote neuroprotection and

to have anti-inflammatory effects, which are thought to indirectly counter two pathologies of AD, the burden of Aβ and the hyperphosphorylation of tau, both being involved in the clinical deficits of AD.

The indication applied for is as add-on therapy for the treatment of early Alzheimer's disease in adults with mild cognitive impairment (MCI) due to Alzheimer's disease or early-stage mild dementia due to Alzheimer's disease, in patients with SIGMAR1 wild-type genotype.

3.1.2. Available therapies and unmet medical need

In the European Union, approved therapies for AD include cholinesterase inhibitors (e.g. donepezil, rivastigmine, galantamine) for patients with mild to moderately severe AD and the non-competitive N-methyl-D-aspartate receptor antagonist memantine for patients with moderate to severe AD. These medications provide modest symptomatic relief but do not address the underlying pathology of the disease (Birks J, 2006).

In November 2024, the CHMP adopted a positive opinion for the monoclonal amyloid beta antibody lecanemab in the treatment of mild cognitive impairment and mild dementia in adults with AD, who are non-carriers or heterozygotes of apolipoprotein E ε4 with confirmed amyloid pathology and Leqembi was authorised by the European Commission on 15 April2025.

In July 2025, CHMP adopted a positive opinion, recommending the granting of a marketing authorisation for the medicinal product Kisunla, intended for the treatment of early symptomatic Alzheimer’s disease in adults who are apolipoprotein E ε4 (ApoE ε4) non-carriers or heterozygotes. The European Commission issued the decision for the approval of Kisunla on 25 September 2025.

However, an unmet medical need for the broad Alzheimer’s disease population is recognised.

3.1.3. Main clinical studies

The blarcamesine clinical development program consists of efficacy and safety data from 5 studies involving a total of 886 participants (22 participants in ANAVEX2-73-001, 32 participants in ANAVEX2-73-002, 24 participants in ANAVEX2-73-003, 508 participants in ANAVEX2-73-AD-004 DB, 300 participants in ANAVEX2-73-AD-EP-004).

A double-blind, placebo-controlled pivotal 48-week study (Study ANAVEX2-73-AD-004), and its long-term extension (Study ANAVEX2-73-AD-EP-004) conducted in patients with early AD constitute the main basis for benefit/risk evaluation.

3.2. Favourable effects

Due to the formal failure of the single pivotal trial, and the complete lack of pre-specification of the analysis methods, no favourable effects can be claimed with certainty.

In the population applied for - SIGMAR1 WT – there was a difference (non-statistically significant in the analysis presented here) in the change from baseline at week 48 for the ADAS-Cog13 in favour of the pooled dose groups of -1.904 (95% CI: -4.036, 0.228). Results for CDR-SB showed a similar trend: difference of -0.532 (95% CI: -1.113, 0.048).

The results on biomarkers, including on the % change in whole brain volume 0.77% (95% CI: 0.29, 1.25, p = 0.0019) are also taken into account.

3.3. Uncertainties and limitations about favourable effects

The dossier does not fully comply with the ICH recommendation and EMA guidelines.

The pharmacokinetic data are incomplete: full characterisation of metabolism, transporter involvement, and human mass-balance studies remain outstanding. This lack of information limits interpretation of both efficacy and safety, as potential active metabolites and drug–drug interactions remain uncharacterised. Overall, internal validity, external reproducibility, and mechanistic plausibility remain uncertain.

The functional co-primary endpoint, ADCS-ADL in the randomised population did not reach statistical significance, rendering the study failed. Other endpoints measuring QoL and caregivers' burden do not show statistically significant results, further questioning that the differences observed (whether or not causally attributable to treatment assignment) may represent patient-relevant benefits.

There is a significant number of issues with respect to pre-specification of the analysis methods (including of the covariates and the covariance structure used in the primary analysis), and multiplicity control between endpoints and doses, that further lower the credibility of any positive claims made.

Overall, due to its methodological flaws, the study presented as pivotal cannot be considered as having established proof-of-concept.

The sample size is considered limited compared to the millions of Alzheimer's disease patients in Europe. Furthermore, the assumption that there will be a 33% dropout rate turned out not to be correct in the case of blarcamesine. The percentages of patients discontinuing due to adverse events were higher in the blarcamesine groups 30 mg and 50 mg compared to the placebo group. This was the case for both the early discontinuations and the ones after week 12. Only 53.3% of the participants in the 50 mg blarcamesine group completed the study, whilst it was 80% of the participants in the case of placebo, who completed the study. The very high rate of discontinuations in the blarcamesine treatment groups adds uncertainty assessment of blarcamesine efficacy, especially when combined with the definition of the analysis population that disproportionately excluded patients assigned to active treatment from the primary analysis.

The duration of the study was too short for an early AD population (only 48 weeks) to show a treatment effect, whilst the recommended minimum duration for a disease modifying treatment is 18 months in the EMA Guideline.

The subgroup analysis on SIGMAR1 WT cannot be considered sufficient to establish efficacy as stemming from a failed trial and lacking replication. Moreover, the results suffer from the same methodological issues highlighted for the main analyses and are not dramatically different from the ITT population.

None of the biomarkers studied can be considered a surrogate: it has not been demonstrated that the presence of an effect as measured by these will necessarily result in clinical benefit.

In conclusion, the efficacy of blarcamesine in the treatment of patients with Alzheimer's disease has not been demonstrated in any Alzheimer's disease population.

3.4. Unfavourable effects

The safety of blarcamesine in the treatment of Early Alzheimer's disease (studies -004 and EP-004) and mild to moderate Alzheimer's disease (studies -002 and -003) in the dose range 10 mg/day to 50 mg/day has been evaluated in four clinical patient studies including 488 individual patients. In the placebo-controlled study -004, blarcamesine was titrated from 10 mg/day over a period of 2 or

3 weeks depending on the protocol version under which patients have been enrolled. Given that the majority of patients have been enrolled under protocol versions > 2 (applying the 3-weeks titration), the safety profile basically reflects this treatment regimen. The two dose groups analysed during the maintenance period up to 48 weeks in study -004 were 30 mg/day and 50 mg/day; nevertheless, the average daily exposure to blarcamesine by the assigned treatment groups was similar and less than 30 mg/day at each of the study visits.

The mean (SD) duration on study drug in study -004 was 225.3 (145.49) days for active total blarcamesine being lower than the 295.6 (97.51) days on placebo. Early study discontinuations were reported twice as high in the active groups (136 patients; 40.2%), and more frequently in the 50 mg/day dose group as compared to 30 mg/day (46.7% vs. 33.7%). The discontinuation rate in the placebo group was 20%.

Long-term safety of blarcamesine has been evaluated in the open-label study EP-004 over a period of up to 144 weeks with 300 rollover patients from either placebo or blarcamesine during study -004. Irrespective of treatment and dose assignment during study -004, all patients underwent a 10-week titration phase starting with blarcamesine 10 mg/day up to 50 mg/day or the best tolerated dose.

The mean (SD) duration on study drug in study EP-004 was 526.36 (371.604) days for the placebo to blarcamesine group and 661.51 (340.824) days for the continued blarcamesine group. More patients previously been treated with placebo during -004 discontinued as compared to patients continuously been treated with blarcamesine from the parent study (66.1% vs. 51.4%), mainly due to AEs.

Common TEAEs with a notably higher incidence in the overall blarcamesine group compared to the placebo group in at least 5% of participants were mainly related to its CNS activity, i.e. dizziness (46.3% vs. 8.9%), confusional state (23.9% vs. 3%), balance disorder (11.6% vs. 1.8%), anxiety (9.9% vs. 3.6%), diarrhoea (9.3% vs. 7.7%), headache (9% vs. 4.8%), fatigue (8.4% vs. 1.2%), lethargy (7.8% vs. 1.8%), somnolence (7.8% vs. 3%), and abnormal dreams (7.5% vs. 0%).

286 of the 324 subjects (85.3%) reported with at least one TEAE in the blarcamesine active total group in study -004 had **treatment-related** (possibly, probably, or definitely related) AEs (versus 51 of 129 subjects [39.5%] with TEAEs in the placebo group). TEAEs from the nervous system disorders SOC, psychiatric disorders SOC, and General disorders and administration site conditions SOC were by majority rated at least possibly related to blarcamesine. Based on the 335 patients in the active total blarcamesine group, there were 44.8%, 21.8%, and 11.4% with treatment-related TEAEs of dizziness, confusional disorder, and balance disorder, respectively.

Reporting of **SAEs** during in study -004 was higher with blarcamesine as compared to placebo (16.7% vs. 10.1%), and higher for the 50 mg/day group as compared to the 30 mg/day group. SAEs most frequently derived from the Nervous System Disorders SOC (e.g. dizziness in 5 [1.5%] patients, 4 of which were rated as related) in line with the safety profile of blarcamesine. Hyponatraemia was reported as SAE in 4 patients in the active total group (no SAE in the placebo group) and more frequently with 50 mg/day as compared to 30 mg/day (3 vs. 1 patient). In study EP-004, 25.7% of patients reported SAEs. The most common SAEs were delirium (2.3%), neuropsychiatric symptoms (2.0%), urinary tract infection (1.3%), and aggression and femoral neck fracture (1.0% each).

108 (32.2%) participants treated with blarcamesine, and 12 (7.1%) participants treated with placebo had at least one **TEAE leading to treatment discontinuation**. The main reasons in the blarcamesine group were dizziness (8.1%) and confusional state (4.2%). Patients more frequently experienced TEAEs leading to treatment discontinuation in the higher dose group. When contrasting the short duration of the titration period (2 to 3 weeks) and the longer duration of the maintenance period (45 to 46 weeks), the relative frequency of discontinuations due to TEAEs was higher during the titration period. Similarly, 24.7% of patients in study EP-004 reported TEAEs leading to treatment

discontinuation, mainly from the psychiatric disorders SOC and the nervous system disorders SOC.

In addition, blarcamesine frequently led to dose changes or drug interruption during study -004 (45.7% and 31.3%), mainly due to nervous system disorders and psychiatric disorders, with a similar incidence in the OLE EP-004.

3.5. Uncertainties and limitations about unfavourable effects

Major uncertainties have been identified questioning the safe use of blarcamesine in the overall AD population as well as in the claimed indication of a subpopulation of AD patients with SIGMAR1 wild-type genotype.

The metabolism of blarcamesine was neither fully characterised in animals, nor in humans. The metabolic conversions determined in rat, dog and human liver microsomal preparations *in vitro* and toxicokinetic data of repeat-dose toxicity studies *in vivo* indicate species-specific metabolic differences. A human mass-balance study is not available, so it remains unclear, if blarcamesine and/or ANAVEX19-144 are excreted unchanged. However, biliary and renal excretion with sex-specific differences were identified in rats. Thus, additional phase 1 and/or phase 2 enzymes are likely involved in the possibly even stereoselective metabolism of blarcamesine.

The evaluation of pharmacokinetic interactions *in vitro* was solely confined to the potential inhibition of CYP isozymes by blarcamesine and its more stable active metabolite ANAVEX19-144 as well as the possible CYP induction by blarcamesine. In addition, a slight contribution of P-gp and BCRP to the gastrointestinal efflux of blarcamesine was identified.

Furthermore, the limited developmental and reproductive toxicology studies indicate that blarcamesine may induce malformations in rats with no or minor safety margins compared to human exposure.

The overall size of the safety database is not acceptable: only 488 *individual* patients have been treated with blarcamesine across four clinical patient studies. With regard to the indicated dose range (blarcamesine 10 mg to30 mg) finally proposed in the label, 263 patients and 206 patients had a treatment exposure duration of at least 6 months and 12 months, respectively, with 102 patients being exposed to the treatment for 24 months or longer. This is out of the specifications indicated in the ICH E1 guideline for drugs intended for the long-term treatment (chronic or repeated intermittent use for longer than 6 months) of non-life-threatening diseases. Moreover, this is clearly insufficient with respect to the disease to-be treated, which is not rare, and - even more important – with respect to the novel mechanism of action claimed by the applicant. So far, no sigma receptor agonist has been approved, which additionally weakens the data package and strongly questions acceptability of one pivotal trial. Despite this, evaluable data were further reduced by the high number of dropouts in study -004 (twice as high in the total active group as compared to placebo; 40.2% vs. 20%) and also in study EP-004 (57%), with the main reason being adverse events questioning the safe use of blarcamesine. This remains of major concern.

The safety profile of blarcamesine that has been insufficiently characterised with regard to its CNS activity. This is concerning against the background of the insufficiently described off-target interactions *in vitro*. With the present knowledge, the receptor profile indicates high affinity of the active metabolite ANAVEX19-144 to the NMDA receptor and the μ-opioid receptor within the range of S1R and muscarinic cholinergic interactions. Blarcamesine consistently induced various neurological impairments in rats and dogs, which coincide with the commonly reported clinical adverse reactions of dizziness, agitation, balance disorders/fall, somnolence, lethargy, fatigue and confusion in human patients. The initial stimulatory effect followed by CNS depression upon prolonged treatment or at higher doses in animals may at least in part be attributable to the presumptive high affinity of

ANAVEX19-144 for the μ-opioid and NMDA receptors. Moreover, the dependence potential of blarcamesine as a new CNS active substance has not been investigated from a nonclinical perspective in accordance with the stepwise approach of the pertinent European guideline (EMEA/CHMP/SWP/94227/2004). Moreover, and as delineated in the *Guideline on the clinical investigation of medicines for the treatment of Alzheimer's disease* (CPMP/EWP/553/95 Rev.2), "*special efforts should be made to assess potential adverse effects that are characteristic of the class of drugs being investigated depending on the action on distinct receptor sites or enzymes (…)*". The same guideline also stipulates that the effect of withdrawal should be systematically monitored.

CNS effects that might be related to dependence potential and withdrawal effects were found clearly increased over placebo and - by majority - also dose-related. These effects might also be related to secondary events, e.g. TEAEs of fall were found increased in the continued blarcamesine group as compared to the placebo to blarcamesine group in study EP-004.

Uncertainty remains with regard to the compilation of the consolidated and longitudinal safety data from studies AD-004 and EP-004.

Uncertainty remains concerning the evaluation of adverse drug reactions: the methodology used to define the ADRs proposed to be included in the product information has not been described. A causality assessment based on temporal aspects and biologic plausibility as well as a reasonable difference to placebo is missing for all TEAEs identified in excess of those observed with placebo.

Uncertainty has been raised with regard to a number of SAEs that have been reported during studies -004 and EP-004, for which a mechanistic rationale remains unclear: Hyponatraemia was reported as SAE in 4 patients on blarcamesine, basically during the first 6 weeks of treatment, more frequently with 50 mg/day as compared to 30 mg/day (3 vs. 1 patient) and possibly related to study drug in 2 patients. However, it could be clarified that the patients had multiple confounders in their medical history/ comedications. Overall, the available data do not support a clear contribution of blarcamesine to these SAEs. SAEs of cerebrovascular accident, UTI, and seizure in EP-004 have been rated as related to blarcamesine. Upon request, a biological plausibility could not be confirmed.

Uncertainty has been raised on TEAEs leading to discontinuation with regard to severity, seriousness, follow-up/ outcome and discussion of narratives for studies -004 and EP-004, especially with regard to the CNS related effects. In a combined analysis of studies -004 and EP-004, 182 patients out of 456 patients (40%) discontinued treatment with blarcamesine, in majority following psychiatric disorders, nervous system disorders, and General disorders and administration site conditions. Dizziness, mainly moderate in severity, led to discontinuation in 7.7% of all patients treated with blarcamesine and had an onset mainly during initiation or following dose increase. All events resolved following discontinuation. Dose reduction attempted in some of the patients prior to discontinuation was obviously insufficient. Confusional state led to discontinuation of blarcamesine in 5% of all patients, was of moderate severity in all but one (severe), and resolved after discontinuation. The onset was not strictly related to treatment initiation. Balance disorder led to discontinuation of blarcamesine in 2.4% of all patients, was often moderate in severity and resolved in all patients after discontinuation. Despite update of section 4.8 of the proposed SmPC, the incidence of TEAEs leading to discontinuation in the description of selected TEAEs should focus on the incidences in study AD-004.

Uncertainty remains on any age-related safety issues, and the safety profile according to other baseline characteristics (intrinsic and extrinsic factors). No data are available for patients with renal or hepatic impairment. While patients with severe renal or hepatic impairment are excluded from treatment with blarcamesine, the lack of data is neither adequately described in the proposed product information (and even contradictory), nor adequately addressed in the proposed RMP.

Pregnancies have not been reported in the blarcamesine clinical programme. However, uncertainty is

raised with respect to the incomplete characterisation of the reproductive and developmental toxicity of blarcamesine, considering the teratogenic effect demonstrated in studies in rats and the lack of a pre- and post-natal development (PPND) study. Although the proposed clinical indication predominantly affects postmenopausal women, it can presently not be ruled out that early cases of dementia in women of childbearing potential will be treated with blarcamesine.

Regarding the insufficient characterisation of the metabolic pathway of blarcamesine, it remains uncertain if ANAVEX19-144 will indeed constitute the only major metabolite in animals and humans. The absence of a qualitative and/or quantitative comparison of metabolic profiles limits the predictive value of toxicity studies in terms of human safety.

In this context, the inadequate characterisation of potential drug interactions is another uncertainty. Although blarcamesine and ANAVEX19-144 did not show clinically relevant direct or time-dependent inhibition of CYP isozymes or induction of CYP1A2, CYP2B6 and CYP3A4 mRNA and enzymatic activities in human hepatocytes, the possible induction of CYP enzymes by the more stable active metabolite ANAVEX19-144 was not tested. Moreover, interactions of both blarcamesine and ANAVEX19-144 with hepatic or renal uptake/efflux transporter were not evaluated. As most AD patients are of advanced age with possibly compromised hepatic and renal clearance and will frequently have to rely on co-medications, the lack of these data constitutes a major risk for the health of the envisaged patient population.

Safety data for CYP or transporter interactions have also not systematically been collected. There were two cases of concomitant use of either a strong CYP3A4 inhibitor (erythromycin) or inducer (carbamazepine) with blarcamesine in study AD-004. No SAEs were reported in these subjects. Safety of blarcamesine in special populations has not been evaluated.

Uncertainty was raised regarding shifts from normal to low in lymphocyte counts during longer treatment exposure with blarcamesine that also led to the reporting of lymphocyte count decreased as TEAEs. However, no biological plausibility or mechanistic rational could be found and it needs to be considered that no effects on white blood cell counts was noted in nonclinical studies. TEAEs associated with increases in serum amylase and lipase in a single patient in the Phase 2 studies rated as related remain uncertain given that these enzymes have not routinely been measured in the clinical programme.

It remains uncertain whether blarcamesine exerts effects on coagulation parameters, since these have not been routinely measured despite being scheduled in the study protocols.

Uncertainty remains with regard to discrete mean decreases in diastolic and systolic blood pressure and slight increases in heart rate up to Week 48 in the 50 mg/day group in study AD-004, supported by data from study EP-004, contrasting preclinical findings pointing towards increases in mean, systolic and diastolic arterial blood pressure in dogs. However, shift analyses were unremarkable.

Lastly, risk factors for the formation of nitrosamines are present and - lacking adequate confirmatory testing – formation of nitrosamines cannot be ruled out.

3.6. Effects Table

Table 32. Effects table for Blarcamesine Anavex in early Alzheimer's disease, in patients with SIGMAR1 wild-type genotype

Effect	Short Description	Unit	Blarcamesine (30 mg/day or 50 mg/day or pooled)	Placebo	Uncertainties/ Strength of evidence	References
Favourable Effects						
ADAS-Cog13	Alzheimer's Disease Assessment Scale-Cognitive Subscale	0 - 85	**LS Mean (SE)** N=139 pooled: 2.981 (0.883)	**LS Mean (SE)** N=83 4.884 (0.859)	UnC: lack of pre-specification of analysis **LS Mean Diff (SE) at 48 weeks from baseline** -1.904 (95% CI: -4.036, 0.228) P = 0.0799	(1)
ADCS-ADL	Alzheimer's Disease Cooperative Study – Activities of Daily Living Scale	0 - 78	**LS Mean (SE)** N=141 pooled: -6.457 (0.760)	**LS Mean (SE)** N=87 -6.929 (0.981)	UnC: Failed co-primary endpoint, lack of pre-specification of analysis **LS Mean Diff (SE) at 48 weeks from baseline** 0.472 (95% CI: -1.974, 2.918) P = 0.7038	(1)
Unfavourable Effects						
Dizziness	Incidence of TEAEs	%	30 mg: 39.5 50 mg: 53.0	8.9		(1)
Balance disorders	Incidence of TEAEs	%	30 mg: 9.6 50 mg: 13.7	1.8		(1)
Lethargy	Incidence of TEAEs	%	30 mg: 7.8 50 mg: 7.7	1.8		(1)
Confusional state	Incidence of TEAEs	%	30 mg: 22.2 50 mg: 25.6	3.0		(1)
Anxiety	Incidence of TEAEs	%	30 mg: 7.8 50 mg: 11.9	3.6		(1)
Depressed mood	Incidence of TEAEs	%	30 mg: 6.0 50 mg: 6.5	2.4		(1)
Fatigue	Incidence of TEAEs	%	30 mg: 8.4 50 mg: 8.3	1.2		(1)
Feeling abnormal	Incidence of TEAEs	%	30 mg: 5.4 50 mg: 4.8	0		(1)

Abbreviations: TEAEs (treatment-emergent adverse events); (1) study ANAVEX2-73-AD-004.

3.7. Benefit-risk assessment and discussion

3.7.1. Importance of favourable and unfavourable effects

The results from the single pivotal double-blind clinical trial AD-004 cannot be considered confirmatory

or supportive of any indication. While the endpoints used cover cognitive and functional aspects, and would have been adequate, had the results been positive and robust in the pre-specified objectives, this is not the case.

On the other hand, a very significant proportion of patients treated with blarcamesine had adverse effects intolerable enough to discontinue treatment. This, in association with the extremely significant uncertainties deriving from a small safety database and several inadequacies of the non-clinical and clinical dossier, establishes that safety has not been demonstrated.

3.7.2. Balance of benefits and risks

The absence of demonstrated efficacy together with the observed safety profile, and the very significant uncertainties on the safety profile, indicate that a positive benefit-risk has not been established. The primary analysis failed. Some nominally statistically significant results were obtained with additional analyses (not always prespecified). Despite the fact that some issues were considered by the applicant (e.g. baseline values as a co-variate), MMRM was not properly pre-specified, in particular with respect to covariance structure and some other covariates that were included. Generally, a major flaw in the study planning and analyses was that analyses were not consistently pre-specified in protocol and SAP and that analyses were changed after study unblinding, which is not in alignment with the basic methodological standards for a confirmatory study.

At every stage of the procedure, the focus of the applicant in the AD populations was different: initially the ITT population, then the post-hoc SIGMAR1/COL24A1 WT Homozygous Population and finally the SIGMAR1 WT Population.

A positive benefit–risk balance for blarcamesine has not been established in the targeted population (or any Alzheimer's disease population).

3.7.3. Additional considerations on the benefit-risk balance

CHMP early dialogue with healthcare professionals and patients

Reaching out to healthcare professional organisations and patient organisations when medicines are in active development will enable CHMP to fully appreciate HPOs the experience and concerns about the management of these conditions and patients' experience and concerns about their conditions. This facilitates understanding aspects that are important for healthcare professionals, such as treatment options, treatment optimisation, unmet medical needs and what benefits they would hope for new treatments. Moreover, from a patient perspective, it helps to understand aspects that are important for patients, such as treatment options, unmet medical needs, quality of life, pregnancy and what benefits they would hope for in new treatments.

Healthcare professional organisations

The European Academy of Neurology (EAN) made some comments for the Anti-Amyloid Monoclonal Antibodies. Drugs such as Lecanemab and Donanemab are designed to target amyloid-beta plaques in the brain, which are thought to contribute to the neurodegenerative process. These treatments have shown some success in reducing amyloid burden, and in reducing the progression of cognitive symptoms. Notably, the high incidence of side effects requires a strict MRI follow up which might engulf the national systems. This represents a huge shift of the therapeutic scenario for AD patients, although caution is important especially for the management of possible side effects, which requires a strict MRI surveillance (Note: this is the exact wording from the Healthcare providers' contribution. However, it is highlighted that approval of Leqembi in EU was issued on 15th April 2025,

https://ec.europa.eu/health/documents/community-register/html/h1891.htm).

The EAN also listed potential benefits and challenges for AD patients of the new medicines.

Benefits of New Drug Candidates

- *Potential to Slow Disease Progression:* Unlike current treatments, new drug candidates target the fundamental causes of the disease, potentially slowing or halting progression. This could significantly improve the quality of life for patients and extend their ability to live independently.
- *Early and Targeted Intervention*: New drugs should ideally be effective when administered in the early stages of the disease, possibly even before clinical symptoms appear. Disease-modifying drugs with biomarkers for patient selection could ensure that individuals at risk for AD receive early intervention to delay progression.
- *Improved Mechanisms of Action:* New drugs work through different pathways which could complement or offer alternatives to amyloid-targeting therapies. By targeting a diverse range of mechanisms, these drugs may be more effective in treating various stages and subtypes of Alzheimer's.
- *Improved Safety and Tolerability*: Safety is a paramount consideration when developing new AD treatments. Many current drugs have side effects such as gastrointestinal issues, dizziness, and more serious risks like brain swelling in anti-amyloid therapies. New medicines should offer better tolerability profiles, with manageable side effects that do not compromise the patient's quality of life.
- *Improved Cognitive and Functional Outcomes*: Ultimately, new treatments should focus on improving patients' cognitive abilities (such as memory, learning, and problem-solving) and functional outcomes (such as the ability to carry out daily activities). A meaningful improvement in daily functioning can significantly impact both the patient's quality of life and the burden on caregivers.

Challenges

However, there are challenges to overcome. Some new drugs have raised concerns regarding their side effects, high cost, and uncertain clinical benefits. Regulatory approvals and long-term data will be necessary to confirm their safety and efficacy. Additionally, the question of accessibility remains critical, as many new drugs could come with high price tags, limiting their availability for many patients who need them.

Acceptable Side Effects and Manageability

Any new medication for AD must balance efficacy with tolerability. AD is a serious brain disorder which is associated with a shortened life expectancy and reduction in quality of life. Therefore, it is possible that patients are willing to risk potentially serious side effects, even perhaps for relatively modest effect sizes. Special emphasis may be needed on initiatives to be put in place to monitor side effects in this patient population due to cognitive impairment and variable insight into symptoms.

In the context of AD treatments, side effects are often unavoidable, but they need to be manageable and not outweigh the benefits.

- *Mild to Moderate Side Effects*: For example, side effects like mild gastrointestinal issues (nausea, diarrhoea) are typically acceptable if they can be managed with adjustments to dosage or supportive care. Similarly, dizziness or headaches may be considered manageable if they are transient or alleviated with proper guidance.

- *Severe or Long-term Side Effects*: On the other hand, severe side effects—such as brain swelling, as seen with some anti-amyloid monoclonal antibodies—are more concerning and would need to be carefully weighed against the therapeutic benefits. If a drug causes significant adverse events that could lead to complications like hospitalization, it may limit the drug's usability, especially in older adults who are already at a higher risk for other health issues.
- *Cognitive Decline and Behavioural Effects*: Another important consideration is the cognitive impact of side effects. If a new drug causes confusion, memory impairment, or worsens the patient's behavioural symptoms (e.g., agitation, hallucinations), it would not be acceptable, given that it would add to the cognitive burden of the disease itself.

Considerations for Pregnant Individuals and People of Child-Bearing Potential

Given the advanced age of most AD patients, pregnancy is generally not a common consideration. However, some genetic cases might interest women with a child-bearing potential, and are some considerations need to be done.

- *Safety of Medications During Pregnancy*: Many AD drugs, especially those that are still under investigation, may not be recommended during pregnancy due to potential risks to the fetus. For example, most cholinesterase inhibitors are classified as pregnancy Category C drugs, meaning that their safety has not been established in pregnancy.
- *Reproductive Risks*: Some newer treatments may have specific contraindications for individuals who are pregnant or planning to become pregnant. For example, therapies targeting amyloid or tau accumulation could have unknown effects on foetal development, particularly if these treatments interfere with cellular processes critical for pregnancy.

Precautions and Counselling: Healthcare providers should carefully evaluate the risks and benefits of any medication for women of child-bearing potential and provide proper counselling. If an individual is pregnant or planning a pregnancy, it may be necessary to discontinue AD treatment or select alternative therapeutic options that are safer for maternal and foetal health.

Patient organisations

Alzheimer Europe conducted three focus group discussions (10 individuals, including people with MCI and dementia) on disease-modifying therapies.

People expressed some disappointment in current treatments, which manage symptoms, though they acknowledged their value in maintaining daily function and the fact that they have minimal side effects. There was strong support for the development of new drugs aimed at halting or slowing disease progression, with hopes for medicines which could support or improve quality of life, with reduced or no side effects.

Concerns were raised over slow drug approval timelines, with participants expressing hope for accelerated access, similar to COVID-19 vaccines. People also highlighted the importance of non-pharmaceutical and preventive interventions, emphasising that these and other treatments should be widely accessible for all people who could benefit. Perspectives on risks and benefits in relation to new treatments varied: some preferred delaying medication due to the risk of side effects, while others were willing to take risks to preserve cognitive function, especially if side effects were likely to be temporary or mild.

Third party interventions

The CHMP received several interventions from third parties, including patients and relatives and caregivers of patients. The interventions were delivered either individually or via organisations or

petitions. Overall, these third parties expressed their views about the epidemiology of Alzheimer disease, the unmet medical need, the desirability of a novel oral treatment for AD that does not present risk of ARIA, and the results of the main study for blarcamesine, including some subgroup analyses (mainly the SIGMAR1/COL24A1 WT homozygous subpopulation) and the biomarker data. Several of the interventions refer to ABCLEAR1 = Alzheimer's Blarcamesine Cognition Efficacy and Resilience gene variant non-carrier population (SIGMAR1 wild type [WT]), ABCLEAR2 = Alzheimer's Blarcamesine Cognition Efficacy and Resilience gene variant non-carrier population (COL24A1 wild type [WT]) and ABCLEAR3 =Alzheimer's Blarcamesine Cognition Efficacy and Resilience gene variants non-carrier population (SIGMAR1 wild type [WT]/COL24A1 wild type [WT]), from the MacFarlane et al (2025) publication. It is noted that the naming of the subgroups is different in this report, as the term "ABCLEAR" has not been used in the majority of the applicant's documentation. The interventions were in support of the approval of blarcamesine.

The CHMP expresses full agreement with the stated unmet need. Furthermore, the CHMP considered those interventions in the context of its assessment and concluded that the observations put forward were already known by CHMP, and, since no new data have been submitted, they had no impact on the CHMP conclusions.

Conditional marketing authorisation

The application is based on a negative pivotal study, whose lack of pre-specification implies results would not in any case have yielded confirmatory value. The trial had a small sample size (compared to the number of AD patients) and was of insufficient duration. The dossier is also incomplete from a non-clinical, clinical pharmacology, and safety perspectives.

As comprehensive data on the product are not available, a conditional marketing authorisation was requested by the applicant during the assessment procedure.

The CHMP considers that the product cannot be recommended for a conditional marketing authorisation as the benefit-risk balance is negative (as discussed), the applicant is unlikely to be able to provide comprehensive data after authorisation, it has not been demonstrated that the product will address an unmet medical need, and the benefits to public health of the immediate availability do not outweigh the risks inherent in the fact that additional data are still required.

- the benefit-risk balance of the medicine is negative

The benefit-risk balance for blarcamesine has not been demonstrated to be favourable in the early Alzheimer's disease population or the subpopulations tested.

- it is unlikely that the applicant will be able to provide comprehensive data post-authorisation

It is not likely that the applicant will be able to provide comprehensive data post-authorisation and a full, detailed and convincing feasibility report was not submitted. The applicant has conducted a single pivotal trial in a small number of patients in a disease, which numbered around 11 million people in Europe and 55 million people worldwide, in 2019. In if a CMA was to be granted for a medicine in AD, it is questioned whether it would be feasible to recruit patients in a post-approval confirmatory study, and to maintain study integrity.

- the medicine does not fulfil an unmet medical need.

Although there is an unmet medical need, blarcamesine does not fulfil it. There is not a clear pharmacological rationale or a mechanistically plausible explanation of the results. The pivotal study failed in the randomised population and the subgroup analyses can be at best considered exploratory.

While the applicant mentioned in their justification that an unmet need persists despite the availability of treatments, a Major Therapeutic Advantage over the existing methods has not been demonstrated.

- the benefit of the medicine's immediate availability to patients is not greater than the risk inherent in the fact that additional data are still required.

Since a favourable benefit risk balance for blarcamesine has not been established, it cannot be assumed that the benefit of immediate availability to patients is greater than the risk inherent in the fact that additional data are still required.

3.8. Conclusions

The overall benefit/risk balance of Blarcamesine Anavex is negative.

4. Recommendations

Outcome

Based on the CHMP review of data on quality, safety and efficacy for Blarcamesine Anavex in the treatment of early Alzheimer's disease in adults with mild cognitive impairment (MCI) due to Alzheimer's disease or early-stage mild dementia due to Alzheimer's disease, in patients with SIGMAR1 wild-type genotype, the CHMP considers by consensus that the quality, safety and efficacy of the above-mentioned medicinal product are not sufficiently demonstrated, and, therefore recommends the refusal of the granting of the conditional marketing authorisation for the above-mentioned medicinal product. The CHMP considers that:

- Quality has not been demonstrated.
  - Risk factors for the formation of nitrosamines are present and – lacking adequate confirmatory testing – formation of nitrosamines cannot be ruled out.
- Efficacy has not been demonstrated.
  - The application is based on a single-pivotal trial that failed to meet its primary objective. The positive results on some of the endpoints claimed by the Applicant, in addition to not being inferentially valid due to the failure of the trial, are further weakened by the lack of pre-specification of several aspects of the statistical methods, and by the amount and handling of missing data.
  - The subgroup analysis presented – in addition to being affected by the amount and handling of missing data and by other methodological issues – cannot be considered of confirmatory value given the failure to establish evidence in the primary analysis population, the limited data supporting a mechanistically plausible rationale and the lack of replication.
- Safety has not been demonstrated.
  - The safety database, with limited exposure and follow-up, does not sufficiently characterise the safety profile of blarcamesine, which is proposed as first-in-class. This is due to the uncertainties in (and extent of) adverse event data collection, as discussed below.
  - There are high treatment discontinuation rates in the main phase (40.2%) and in the extension (57.3%) of the pivotal study, mainly due to CNS-related adverse events that raise significant tolerability concerns.
  - The characterisation of the safety profile is inadequate, with insufficient evaluation of CNS-

related effects such as dependence or withdrawal symptoms, and with no adequate integrated long-term safety analysis.

- Further critical deficiencies in the demonstration of safety derive from non-clinical data pointing to malformations in reproductive toxicity studies, from the insufficiently justified postponement of carcinogenicity testing, from the inadequate drug-drug interaction studies and from the missing characterisation of metabolic profiles including human mass-balance evaluation.

As efficacy and safety have not been demonstrated, the benefit-risk cannot be considered positive, the medicine would not fulfil an unmet need, and there is no benefit of the medicine's immediate availability that could outweigh the risks inherent in the fact that additional data are still required. Furthermore, it is not considered likely that – in case of approval – the applicant would be able to provide comprehensive data post-authorisation. Hence, a Conditional Marketing Authorisation cannot be granted.

Due to the aforementioned concerns a satisfactory summary of product characteristics, labelling, package leaflet, pharmacovigilance system, risk management plan and post-authorisation measures to address other concerns as outlined in the list of outstanding issues cannot be agreed at this stage.

New Active Substance Status

Based on the CHMP review of the available data, the CHMP considers that Blarcamesine is to be qualified as a new active substance in itself, as it is not a constituent of a medicinal product previously authorised within the European Union.