

Report on the Deliberation Results

December 3, 2025

Pharmaceutical Evaluation Division, Pharmaceutical Safety Bureau
Ministry of Health, Labour and Welfare

Brand Name	Zurzuvae Capsules 30 mg
Non-proprietary Name	Zuranolone (JAN*)
Applicant	Shionogi & Co., Ltd.
Date of Application	September 27, 2024

Results of Deliberation

In its meeting held on October 30, 2025, the First Committee on New Drugs concluded that the product may be approved and that this result should be presented to the Pharmaceutical Affairs Council.

The product is not classified as a biological product or a specified biological product. The re-examination period is 8 years. The drug product is classified as a powerful drug, and its drug substance is classified as a poisonous drug.

Approval Condition

The applicant is required to develop and appropriately implement a risk management plan.

The Committee members made the following comments on precautions and information concerning the indications and dosage and administration in the package insert:

- In a phase III study that evaluated the add-on effect of zuranolone to other antidepressants, the response rate tended to be lower in the zuranolone group than in the placebo group, and the study failed to demonstrate the efficacy of zuranolone as an add-on to other antidepressants. This information should be described in the package insert.
- The statements in the PRECAUTIONS CONCERNING INDICATIONS section should be clarified further to avoid misunderstanding about the scope of eligible patients.
- The package insert should make it clear that re-treatment with zuranolone is one of the available options, so that repeated dosing of zuranolone will be considered more carefully.

Taking account of the above, the First Committee on New Drugs concluded that the package insert should incorporate the Committee members' comments to provide precautions and information.

As a result of this conclusion, there is no change to the results of the review.

**Japanese Accepted Name (modified INN)*

This English translation of this Japanese review report is intended to serve as reference material made available for the convenience of users. In the event of any inconsistency between the Japanese original and this English translation, the Japanese original shall take precedence. PMDA will not be responsible for any consequence resulting from the use of this reference English translation.

Review Report

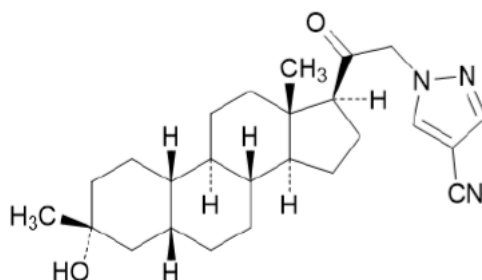
October 8, 2025

Pharmaceuticals and Medical Devices Agency

The following are the results of the review of the following pharmaceutical product submitted for marketing approval conducted by the Pharmaceuticals and Medical Devices Agency (PMDA).

Brand Name	Zurzuvae Capsules 30 mg
Non-proprietary Name	Zuranolone
Applicant	Shionogi & Co., Ltd.
Date of Application	September 27, 2024
Dosage Form/Strength	Hard capsules: Each hard capsule contains 30 mg zuranolone.
Application Classification	Prescription drug, (1) Drug with a new active ingredient

Chemical Structure



Molecular formula: C₂₅H₃₅N₃O₂

Molecular weight: 409.56

Chemical name: 1-(3α-Hydroxy-3β-methyl-20-oxo-19-nor-5β-pregnan-21-yl)-1H-pyrazole-4-carbonitrile

Items Warranting Special Mention None

Reviewing Office Office of New Drug III

Results of Review

On the basis of the data submitted, PMDA has concluded that the product has efficacy in the treatment of depression and depressive state, and that the product has acceptable safety in view of its benefits (see Attachment).

As a result of its review, PMDA has concluded that the product may be approved for the indications and dosage and administration shown below, with the following condition. The product is not classified as a biological

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product or a specified biological product. The drug product is classified as a powerful drug, and its drug substance is classified as a poisonous drug.

Indications

Depression and depressive state

Dosage and Administration

The usual adult dosage is 30 mg zuranolone taken orally once daily after the evening meal for 14 days. The treatment may be repeated after a minimum interval of 6 weeks from the end of dosing.

Approval Condition

The applicant is required to develop and appropriately implement a risk management plan.

Review Report (1)

August 29, 2025

The following is an outline of the data submitted by the applicant and content of the review conducted by the Pharmaceuticals and Medical Devices Agency (PMDA).

Product Submitted for Approval

Brand Name	Zurzuvae Capsules 30 mg
Non-proprietary Name	Zuranolone
Applicant	Shionogi & Co., Ltd.
Date of Application	September 27, 2024
Dosage Form/Strength	Hard capsules: Each hard capsule contains 30 mg zuranolone.
Proposed Indications	Depression and depressive state

Proposed Dosage and Administration

The usual adult dosage is 30 mg zuranolone taken orally once daily after the evening meal for 14 days. The treatment may be repeated at least 6 weeks after the end of dosing.

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List of Abbreviations

See Appendix.

1. Origin or History of Discovery, Use in Foreign Countries, and Other Information

Zuranolone is an analog of a neuroactive steroid allopregnanolone and was discovered by Sage Therapeutics¹⁾ (the US). It is expected to exert antidepressant effects by binding at an allosteric site on γ -aminobutyric acid type A (GABA_A) receptors and enhancing GABA_A receptor function.

In June 2018, Sage Therapeutics granted the applicant exclusive rights in Japan, Taiwan, and South Korea to develop and market zuranolone. In Japan, the applicant initiated a clinical study in 2020 and has now filed a marketing application for zuranolone based on the results from a Japanese phase III study etc.

Outside Japan, zuranolone was approved for postpartum depression in the US in August 2023. As of August 2025, zuranolone has been approved in the UK, and the EU application is under review. Zuranolone has not been approved for major depressive disorder (MDD) in any country or region.

2. Quality and Outline of the Review Conducted by PMDA

2.1 Drug substance

2.1.1 Characterization

The drug substance is a white powder or masses. Its appearance, solubility, melting point, optical rotation, partition coefficient, hygroscopicity, thermal analysis (differential scanning calorimetry, thermogravimetric analysis), and crystalline polymorphism have been determined. Although at least 15 polymorphic forms of the drug substance (Forms 1 to 15) have been identified, only Form 1 (anhydrous form) is produced by the proposed commercial manufacturing process and has been demonstrated to be stable at room temperature.

Its chemical structure has been elucidated by elemental analysis, mass spectrometry, nuclear magnetic resonance spectroscopy (NMR) (¹H-NMR, ¹³C-NMR), infrared absorption spectroscopy (IR), ultraviolet-visible spectroscopy (UV-VIS), X-ray crystallography, and X-ray powder diffraction. Zuranolone has 8 chiral centers, and appropriate controls are implemented to ensure that the desired optical isomer is selectively obtained.

2.1.2 Manufacturing process

The drug substance is synthesized using Starting Material A and Starting Material B.

A quality control strategy was developed based on the following etc. (Table 1).

- Identification of critical quality attributes (CQAs)
- Identification of critical process parameters (CPPs) through quality risk assessment, design of experiments, etc.

1) Sage Therapeutics has been acquired by Supernus Pharmaceuticals.

Table 1. Overview of drug substance quality control strategy

CQA	Method of control
Content	Specification
Appearance	Specification
Identity	Specification
██████████	Manufacturing process, Specification
Related substances	Manufacturing process, Specification
Residual solvents	Manufacturing process, Specification
Water content	Specification
Inorganic impurities	Specification
██████████	Manufacturing process, Specification
██████████	Manufacturing process
██████████	Manufacturing process

Steps ██████████, ██████████, ██████████, and ██████████ have been defined as critical steps. Compound A, Compound B, and Compound C are controlled as critical intermediates.

2.1.3 Control of drug substance

The proposed specifications for the drug substance consist of content, appearance, identity (IR, high performance liquid chromatography [HPLC]), purity (██████████ (██████████)), related substances (HPLC), residual solvents (gas chromatography [GC]), water content, residue on ignition, ██████████, and assay (HPLC).

2.1.4 Stability of drug substance

The primary stability studies on the drug substance are shown in Table 2. The stability results indicated that the drug substance is stable. Photostability data showed that the drug substance is photostable.

Table 2. Stability studies on drug substance

Study	Primary batches	Temperature	Humidity	Storage package	Storage period
Long-term	3 commercial-scale batches	25°C	60%RH	██████████ polyethylene bag +	24 months
Accelerated		40°C	75%RH	██████████ drum	6 months

Based on the above, a re-test period of ██████████ months was proposed for the drug substance when stored in a ██████████ polyethylene bag within a ██████████ drum at room temperature. The long-term testing will be continued up to ██████████ months.

2.2 Drug product

2.2.1 Description and composition of drug product and formulation development

The drug product is an immediate-release hard capsule containing 30 mg zuranolone and the following excipients: silicified microcrystalline cellulose, D-mannitol, croscarmellose sodium, colloidal silicon dioxide, and sodium stearyl fumarate.

2.2.2 Manufacturing process

The drug product is manufactured through a process comprised of ██████████/blending, ██████████, ██████████,

encapsulation, filling/packaging/labeling, and testing/storage. Encapsulation has been defined as a critical step. Process control items and values have been established for encapsulation and filling/packaging/labeling.

A quality control strategy was developed based on the following etc. (Table 3).

- Identification of CQAs
- Identification of material attributes that impact CQAs, determination of the proven acceptable ranges (PARs) for manufacturing process parameters

Table 3. Overview of drug product quality control strategy

CQA	Method of control
Strength	Manufacturing process, Specification
Appearance	Specification
Related substances	Specification
	Specification
Uniformity of dosage units	Manufacturing process, Specification
	Specification
Dissolution	Manufacturing process, Specification

2.2.3 Control of drug product

The proposed specifications for the drug product consist of strength, appearance, identity (), purity [related substances (HPLC)], , uniformity of dosage units [], , dissolution (HPLC), and assay (HPLC). Purity [related substances (HPLC)] was added in the drug product specification in the course of regulatory review.

2.2.4 Stability of drug product

The primary stability studies on the drug product are shown in Table 4. The stability results indicated that the drug product is stable. Photostability data showed that the drug product is photostable.

Table 4. Stability studies on drug product

Study	Primary batches	Temperature	Humidity	Storage package	Storage period
Long-term	3 commercial-scale batches	25°C	60%RH	Blister pack	36 months
Accelerated		40°C	75%RH		6 months

Based on the above, in accordance with the "Guideline on Evaluation of Stability Data" (PMSB/ELD Notification No. 0603004 dated June 3, 2003) (ICH Q1E guideline), a shelf-life of 48 months was proposed for the drug product when packaged in a blister pack () and () and stored at room temperature. The long-term testing will be continued up to months.

2.R Outline of the review conducted by PMDA

Based on the submitted data, PMDA concluded that the quality of the drug substance and the drug product is adequately controlled.

3. Non-clinical Pharmacology and Outline of the Review Conducted by PMDA

The applicant submitted the results from the following non-clinical pharmacology studies of zuranolone: primary pharmacodynamic, secondary pharmacodynamic, safety pharmacology, and pharmacodynamic drug

interaction studies. In some studies, the metabolites of zuranolone in humans,⁷⁾ i.e., M117, M125, M135, and M136 were also evaluated. The main study results are described below. Unless otherwise specified, 15% hydroxypropyl- β -cyclodextrin (HP β CD) aqueous solution was used as vehicle in *in vivo* studies.

3.1 Primary pharmacodynamics

3.1.1 *In vitro* studies

3.1.1.1 Binding to GABA_A receptor

The effects of zuranolone on ³⁵S-ligand binding to GABA_A receptor Cl⁻ channels in rat cerebrocortical membranes were evaluated. Zuranolone inhibited the ligand binding in a concentration-dependent manner, with inhibitory constant (K_i) values of 3.9 to 6.0 nmol/L (CTD 4.2.1.1-01, 4.2.1.1-02). The metabolites of zuranolone, M117, M125, M135, and M136 were also evaluated. M117, M135, and M136 inhibited the ligand binding in a concentration-dependent manner, with K_i values of 140 nmol/L, 2800 nmol/L, and 2200 nmol/L, respectively, and M125 did not inhibit the ligand binding at the concentrations up to 10 μ mol/L (CTD 4.2.1.1-03).

3.1.1.2 Effects on GABA-evoked currents at GABA_A receptor subunit combinations $\alpha_1\beta_2\gamma_2$ and $\alpha_4\beta_3\delta$

The effects of zuranolone on GABA (2-10 μ mol/L)-evoked Cl⁻ currents were examined using a whole-cell patch-clamp technique in cells (Ltk cells [mouse fibroblast cell line], human embryonic kidney 293 [HEK293] cells, or Chinese hamster ovary [CHO] cells) expressing recombinant human GABA_A receptor subtypes $\alpha_1\beta_2\gamma_2$ and $\alpha_4\beta_3\delta$, which are the most abundant postsynaptic and extrasynaptic GABA_A receptors in the brain, respectively. The results are shown in Table 5. Zuranolone potentiated GABA-evoked currents at both GABA_A receptor subtypes in a concentration-dependent manner.

Table 5. Enhancement of recombinant human $\alpha_1\beta_2\gamma_2$ or $\alpha_4\beta_3\delta$ GABA_A receptor function by zuranolone

Receptor type	Cell type	GABA concentration (μ mol/L)	EC ₅₀ (nmol/L)	E _{max} (%)	Patch-clamp method	CTD
$\alpha_1\beta_2\gamma_2$	Ltk	2	178-701	279-706	Automated	4.2.1.1-04
$\alpha_1\beta_2\gamma_2$	Ltk	2	294-1011	587-694	Manual	4.2.1.1-07
$\alpha_1\beta_2\gamma_2$	Ltk	2	374	1041	Manual	4.2.1.1-08
$\alpha_1\beta_2\gamma_2$	HEK293	4	587	2049	Automated	4.2.1.1-05
$\alpha_1\beta_2\gamma_2$	HEK293	4	732	2008		
$\alpha_4\beta_3\delta$	HEK293	10	250	3912		
$\alpha_4\beta_3\delta$	HEK293	10	116	4051		
$\alpha_4\beta_3\delta$	CHO	2	140-737	226-686	Manual	4.2.1.1-06
$\alpha_4\beta_3\delta$	CHO	2	163	639	Manual	4.2.1.1-08

The effects of zuranolone (0.01, 0.1, 1, or 10 μ mol/L) on steady-state currents in the presence of GABA (2 μ mol/L) were examined in CHO cells expressing GABA_A receptors containing $\alpha_4\beta_3\delta$ subunits. Zuranolone potentiated GABA-evoked steady-state currents in a concentration-dependent manner, suggesting that zuranolone potentiates GABA-evoked currents even during persistent activity (CTD 4.2.1.1-09).

7) Among the metabolites of zuranolone identified in a mass balance study (CTD 5.3.3.1-02, 5.3.2.2-12), zuranolone epimer M117, M125, which had a plasma AUC_{0-last} greater than zuranolone (N-des(cyanopyrazole)-dihydroxy-zuranolone carboxylic acid [zuranolone epimer]), and M135 and M136, which were less polar than zuranolone and had abundance at >25% of the AUC_{0-last} of zuranolone in plasma (both dehydrated-zuranolone) were evaluated.

The effect of zuranolone (200 nmol/L) on Cl⁻ currents evoked by piperidine-4-sulfonic acid, which is a partial agonist at GABA_A receptors and is therefore less likely than GABA to induce desensitization of the receptor, was examined in Ltk cells expressing $\alpha_1\beta_2\gamma_2$ GABA_A receptors. The concentration-response of piperidine-4-sulfonic acid was measured with and without zuranolone at $\alpha_1\beta_2\gamma_2$ GABA_A receptors. Data were normalized to the maximum current evoked by a saturating concentration (100 μ mol/L) of GABA. When zuranolone was applied in addition to piperidine-4-sulfonic acid, the 50% effective concentration (EC₅₀) of piperidine-4-sulfonic acid shifted from 47 μ mol/L to 25 μ mol/L, and the E_{max} of piperidine-4-sulfonic acid shifted from 24% to 50%. Zuranolone potentiated the currents evoked by piperidine-4-sulfonic acid (CTD 4.2.1.1-10).

3.1.1.3 Effects on human GABA_A receptors composed of various subunits

The effects of zuranolone on GABA-evoked Cl⁻ currents were examined in cells (Ltk cells, HEK293 cells, or CHO cells) expressing recombinant human GABA_A receptor subtypes other than $\alpha_1\beta_2\gamma_2$ and $\alpha_4\beta_3\delta$. The results are shown in Table 6. Zuranolone potentiated GABA-evoked currents at all GABA_A receptor subtypes tested in a concentration-dependent manner.

Table 6. Enhancement of the function of recombinant human GABA_A receptors composed of various subunits by zuranolone

Receptor type	Cell type	GABA concentration (μ mol/L)	EC ₅₀ (nmol/L)	E _{max} (% potentiation)	Patch-clamp method	CTD
$\alpha_2\beta_2\gamma_2$	Ltk	2	NC ^{a)}	NC ^{a)}	Automated	4.2.1.1-11
$\alpha_2\beta_2\gamma_2$	Ltk	2	184	1344	Manual	4.2.1.1-12
$\alpha_1\beta_3\gamma_2$	HEK293	5	390	712	Automated	4.2.1.1-13
$\alpha_2\beta_3\gamma_2$	HEK293	2	516	1060	Automated	
$\alpha_3\beta_3\gamma_2$	HEK293	12.5	293	341	Automated	
$\alpha_5\beta_3\gamma_2$	HEK293	2	298	564	Automated	
$\alpha_6\beta_3\delta$	CHO	1	147	702	Manual	4.2.1.1-14
$\alpha_6\beta_3\delta$	CHO	1	68	203	Manual	4.2.1.1-15

a) EC₅₀ or E_{max} was not calculated. Zuranolone at 1 and 10 μ mol/L potentiated GABA (2 μ mol/L)-evoked currents by 184% and 462%, respectively.

3.1.1.4 Direct gating of GABA_A receptors

The effects of zuranolone on Cl⁻ currents in the absence of GABA were examined using a whole-cell patch-clamp technique in Ltk cells expressing human GABA_A receptors containing $\alpha_1\beta_2\gamma_2$ subunits and CHO cells expressing human GABA_A receptors containing $\alpha_4\beta_3\delta$ subunits. At both receptor subtypes, zuranolone (10 μ mol/L) produced <50% of the current evoked by GABA (2 μ mol/L) (Reference data CTD 4.2.1.4-06, 4.2.1.1-17).

The effects of zuranolone on Cl⁻ currents were examined using a two-electrode voltage clamp technique in *Xenopus laevis* oocytes expressing human GABA_A receptors containing $\alpha_1\beta_2\gamma_2$ or $\alpha_4\beta_3\delta$ subunits. Zuranolone (1-100 μ mol/L) exhibited direct gating activity at human $\alpha_4\beta_3\delta$ GABA_A receptors, but its direct gating effect was weaker than its positive allosteric modulator (PAM) effect. Zuranolone exhibited no direct gating activity at human $\alpha_1\beta_2\gamma_2$ GABA_A receptors (Reference data CTD 4.2.1.1-18).

3.1.1.5 Effects on tonic and phasic GABA currents

The effects of zuranolone (0.01, 0.03, 0.1, 0.3, 1, or 3 μ mol/L) on tonic GABA currents (holding currents) and phasic GABA currents (inhibitory postsynaptic currents [IPSCs]) were examined using a whole-cell patch-clamp technique in rat cerebellar granule cells and dentate gyrus granule cells. In cerebellar granule cells,

zuranolone (1-3 $\mu\text{mol/L}$) increased the amplitude of holding currents, and zuranolone (3 $\mu\text{mol/L}$) prolonged IPSC decay (CTD 4.2.1.1-19). In dentate gyrus granule cells, zuranolone (1-3 $\mu\text{mol/L}$) increased the amplitude of holding currents, and zuranolone (0.3-3 $\mu\text{mol/L}$) prolonged IPSC decay (CTD 4.2.1.1-20).

3.1.1.6 Sustained increase in tonic and phasic GABA currents

Hippocampal slices from mice were pre-incubated with zuranolone (0.1 or 1 $\mu\text{mol/L}$) for 15 minutes and then perfused for ≥ 30 minutes to wash out zuranolone. Tonic GABA currents were measured from dentate gyrus granule cells, and current density was determined. Current density increased after pre-incubation with zuranolone (1 $\mu\text{mol/L}$) compared to vehicle-treated cells (Reference data CTD 4.2.1.1-21).

Rat brain slices were pre-incubated with zuranolone (1 $\mu\text{mol/L}$) for 15 minutes and then perfused for 45 minutes to wash out zuranolone, and tonic and phasic GABA currents were measured from dentate gyrus granule cells. Pre-incubation with zuranolone increased the amplitude of tonic and phasic GABA currents, but did not affect IPSC decay time (Reference data CTD 4.2.1.1-22).

3.1.2 In vivo studies

3.1.2.1 Study in mouse model of depression induced by social defeat stress (CTD 4.2.1.1-46)

Vehicle or zuranolone (1 mg/kg) was administered once daily for 7 days in mice with depression induced by social defeat stress (C57BL/6J mice),⁸⁾ and the effect of zuranolone on the interaction time with ICR mice in a social interaction test was assessed. In C57BL/6J mice, zuranolone increased the interaction time with ICR mice, compared with vehicle, on Day 1 through Day 7.

3.1.2.2 EEG studies in rats (Reference data CTD 4.2.1.1-34, 4.2.1.1-35)

Vehicle (30% [w/v] sulfobutylether- β -cyclodextrin) or zuranolone was administered intravenously (1, 3, or 5 mg/kg) or orally (0.3, 3, or 20 mg/kg) in rats, and the effects of zuranolone on electroencephalography (EEG) power spectrum were assessed. Zuranolone produced dose-dependent increases in the δ -, θ -, α -, β -, and γ_1 -band EEG power via either route of administration, and the minimum effective dose of zuranolone at which an increase in the β -band EEG power (a biomarker of GABA receptor PAM activity) was observed within 1 hour post-dose was 1 mg/kg by intravenous route and 3 mg/kg by oral route.

3.2 Secondary pharmacodynamics

3.2.1 Effects on various receptors, transporters, etc.

The effects of zuranolone (10 $\mu\text{mol/L}$) on ligand binding were evaluated in various assays against 72 receptors including the GABA_A receptor, 4 transporters, and 2 enzymes. Zuranolone caused $\geq 50\%$ inhibition of ligand binding at the following receptors, except for the GABA_A receptor: the sigma receptor (87%-89% inhibition) and the glycine receptor (1.2%-57% inhibition). In these assays, the functional activity of zuranolone on 3 receptors and 2 enzymes was assessed. Zuranolone exhibited transient receptor potential cation channel

8) Each C57BL/6J mouse was subjected to aggression by placing the mouse into the cage of a novel ICR mouse for 10 to 15 minutes each day for 10 days. Then, a social interaction test was performed in an open field chamber outfitted with a wire mesh cage at one end. C57BL/6J mice were selected if they spent less time in the zone around the cage with vs. without an ICR mouse.

subfamily V member 1 (TRPV1) agonist activity, and the concentration-response of zuranolone for TRPV1 was measured. Zuranolone exhibited apparent functional antagonist activity at TRPV1 ($IC_{50} = 12 \mu\text{mol/L}$) (CTD 4.2.1.2-01, 4.2.1.2-02, Reference data CTD 4.2.1.2-03).

The effects of zuranolone ($10 \mu\text{mol/L}$) on ligand binding to 15 receptors that are generally known as the target for dependence-producing drugs, were assessed. Zuranolone inhibited ligand binding to the sigma 2 receptor by $\geq 50\%$ (85% inhibition) (Reference data CTD 4.2.1.2-04).

3.2.2 Effects on nuclear receptors (CTD 4.2.1.2-05)

The functional activity of zuranolone (0.4 , 2 , or $10 \mu\text{mol/L}$) on 22 nuclear receptors was assessed by a luciferase assay. Zuranolone ($10 \mu\text{mol/L}$) produced a 2.3-fold increase in pregnane X receptor activation compared with vehicle (dimethyl sulfoxide [DMSO]). Zuranolone did not exhibit $\geq 50\%$ inhibition at any receptor relative to vehicle.

3.2.3 Effects in various seizure models

Table 7 shows the results of studies that evaluated the effects of zuranolone in various seizure models.

Table 7. Overview of studies that evaluated the effects of zuranolone in various seizure models

Model	Animal species	Doses (mg/kg)	Route of administration	Noteworthy findings	CTD
Model of pentylenetetrazol-induced seizures	Mouse	0, 1, 3, 10	Oral	Zuranolone (0.3 - 10 mg/kg) increased the latency to tonic seizures.	Reference data 4.2.1.1-23
		0, 0.1, 0.3, 1	Oral		Reference data 4.2.1.1-24
Model of 6-Hz electroshock-induced seizures	Mouse	0, 0.3, 1, 3	Intraperitoneal	Zuranolone prevented clonic seizures ($ED_{50} = 1.897 \text{ mg/kg}$).	Reference data 4.2.1.1-27
Fmr1-knockout model	Mouse	0, 1, 5	Intraperitoneal	Zuranolone (1 - 5 mg/kg) blocked audiogenic seizures.	Reference data 4.2.1.1-28
Model of kainic acid-induced mesial temporal lobe epilepsy	Mouse	0, 1, 3, 5	Intraperitoneal	Zuranolone (1 - 5 mg/kg) reduced hippocampal paroxysmal discharges.	Reference data 4.2.1.1-30
Lithium-pilocarpine-induced status epilepticus model	Rat	0, 1, 3, 5	IV	Zuranolone (3 - 5 mg/kg) reduced EEG power.	Reference data 4.2.1.1-31
Refractory status epilepticus model	Rat	0, 0.3, 1, 2, 3, 4, 5	IV	Zuranolone (1 - 5 mg/kg) reduced EEG power.	Reference data 4.2.1.1-32 Reference data 4.2.1.1-33

3.2.4 Anxiolytic/anti-conflict effects

Zuranolone 0.3 , 1 , or 3 mg/kg was administered intraperitoneally to mice, and the elevated plus maze test was performed at 30 minutes post-dose. In the 5-minute session, zuranolone at 1 and 3 mg/kg increased the number of entries and the time spent in the open arms of the elevated plus maze compared to vehicle (Reference data CTD 4.2.1.1-36).

Zuranolone 0.3 , 1 , or 3 mg/kg was administered intraperitoneally to rats, and the elevated plus maze test was performed at 60 minutes post-dose. In the 5-minute session, zuranolone did not change the number of entries or the time spent in the open arms compared to vehicle (Reference data CTD 4.2.1.1-37).

Zuranolone 0.5 , 1 , 2.5 , or 5 mg/kg was administered intraperitoneally to rats, and the Vogel conflict test was

performed. Increases in the total number of licks and electric shocks were observed in the zuranolone 2.5 and 5 mg/kg groups compared with the vehicle group at 30 minutes post-dose, but there were no effects of zuranolone at 60 minutes post-dose (Reference data CTD 4.2.1.1-38 to 4.2.1.1-40).

3.2.5 Effect on oral dyskinesia (Reference data CTD 4.2.1.1-41)

While reserpine-induced oral dyskinesia was not modified by an agonist of γ subunit-containing GABA_A receptors, i.e., diazepam, an agonist of δ subunit-containing GABA_A receptors, gaboxadol (THIP), and baclofen, which potentiates tonic GABA currents mediated by extrasynaptic GABA_A receptors (*J Neurosci.* 2013; 33: 3780-5, *J Neurosci.* 2013; 33: 3738-43), abolished the manifestation of reserpine-induced oral dyskinesia (*Behav Brain Res.* 2005; 160: 51-9). Given these findings, the effect of zuranolone (0.3-3 mg/kg, intraperitoneal injection) on reserpine (1 mg/kg, subcutaneous injection)-induced oral dyskinesia was evaluated in rats. Zuranolone 3 mg/kg reduced tongue protrusion and vacuous chewing movements induced by reserpine.

3.2.6 Effects on spontaneous locomotion activity and motor coordination

Zuranolone (1-10 mg/kg) was administered intraperitoneally to mice, and the effects of zuranolone on spontaneous locomotor activity were evaluated. Zuranolone at 5 to 10 mg/kg tended to decrease the total distance travelled in an open field chamber (Reference data CTD 4.2.1.1-42, 4.2.1.1-43).

Zuranolone (1-10 mg/kg) was administered intraperitoneally to mice, and the effect of zuranolone on motor coordination was evaluated in a rotarod performance test. The latency to drop off the rotarod tended to be reduced at 10 mg/kg of zuranolone (Reference data CTD 4.2.1.1-44).

3.3 Safety pharmacology

An overview of safety pharmacology studies is shown in Table 8.

Table 8. Overview of safety pharmacology studies

Organ systems evaluated	Test system	Endpoints/ Method of assessment, etc.	Doses or concentrations	Route of administration	Findings	CTD
CNS	Rat (10 males/group)	Clinical observations, FOB	0, ^{a)} 3, 10, 22.5 mg/kg	Oral	(Clinical observations) ≥10: lack of righting reflex 22.5: decreased activity, ataxia, hypersensitivity to touch and/or sound (FOB) ≥3: palpebral closure, abnormal gait ≥10: lack of righting reflex 22.5: decreased general arousal, increased hindlimb splay, increased thermal response latency, impaired posture, decreased activity	4.2.1.3-01
Cardiovascular system	HEK293 cells or CHO cells	Cardiac ion channel currents ^{b)}	0, ^{c)} 0.01, 0.03, 0.1, 0.3, 1, 3, 10, 30 µmol/L	<i>In vitro</i>	IC ₅₀ >30 µmol/L	Reference data 4.2.1.3-02
	HEK293 cells	hERG channel currents	0, ^{d)} 0.1, 0.3, 1, 3 µmol/L	<i>In vitro</i>	IC ₅₀ >3 µmol/L	4.2.1.3-03
	Dog (4 males)	Clinical observations, blood pressure, heart rate, ECG (Conscious, telemetry)	0, ^{e)} 0.4, 1, 2.5 mg/kg	Oral	(Clinical observations) ≥1: circling, decreased activity, eyelids partially or completely closed, lateral recumbency, ataxia, salivation, shallow/rapid breathing, tremors 2.5: decreased body temperature (Cardiovascular parameters) ≥1: decreased systolic blood pressure, decreased mean blood pressure, slight increase in diastolic blood pressure, slight increase in mean arterial pressure, increased heart rate (ECG) 2.5: decreased RR interval, decreased PR interval, decreased QT interval	4.2.1.3-04
Respiratory system	Rat (8 males/group)	Clinical observations, respiratory rate, tidal volume, minute volume (whole body plethysmography)	0, ^{a)} 3, 10, 22.5 mg/kg	Oral	(Clinical observations) ≥10: decreased activity, ataxia, lateral recumbency, prostration, red material around the eyes, eyelids partially or completely closed (Respiratory system) ≥3: increased tidal volume, slight increase in minute volume 22.5: slight decrease in respiratory rate	4.2.1.3-05

a) Vehicle: PEG-8 caprylic/capric glycerides

b) Nav1.5, hERG, KCNQ1/minK, Cav1.2, Kv4.3/KChIP2.2, Kv1.5, Kir2.1, and HCN2

c) Vehicle: 0.3% DMSO

d) Vehicle: saline, 0.3% DMSO

e) Vehicle: 30% HPβCD aqueous solution

3.4 Pharmacodynamic drug interactions

3.4.1 Effects on GABA-evoked currents (CTD 4.2.1.1-06, 4.2.1.1-07, 4.2.1.4-01 to 4.2.1.4-05)

In Ltk cells expressing human $\alpha_1\beta_2\gamma_2$ GABA_A receptors or CHO cells expressing human $\alpha_4\beta_3\delta$ GABA_A receptors, the modulatory effects of zuranolone (0.01-10 µmol/L) in combination with diazepam (0.1 µmol/L), pentobarbital (300 µmol/L), or propofol (0.1 or 3 µmol/L) on GABA-evoked currents, in the presence of GABA (2 µmol/L), were examined using a whole-cell patch-clamp technique, for drug interaction evaluations. The results are shown in Table 9 and Table 10. Compared with zuranolone alone (Table 5), zuranolone in combination with diazepam tended to decrease the EC₅₀ and increase the E_{max} at $\alpha_1\beta_2\gamma_2$ GABA_A receptors. The presence of pentobarbital tended to increase the E_{max} at both receptor subtypes. The presence of propofol tended to reduce the E_{max} at $\alpha_1\beta_2\gamma_2$ GABA_A receptors and increase the EC₅₀ at $\alpha_4\beta_3\delta$ GABA_A receptors.

Table 9. Enhancement of human $\alpha_1\beta_2\gamma_2$ GABA_A receptor function by zuranolone + other GABA_A receptor modulators

Treatment group	EC ₅₀ (nmol/L)	E _{max} (%)	CTD
Zuranolone + Diazepam	112	1162	4.2.1.4-01
Zuranolone + Pentobarbital	465	1407	4.2.1.4-03
Zuranolone + Propofol (0.1 μ mol/L)	920	317	4.2.1.4-04

Table 10. Enhancement of human $\alpha_4\beta_3\delta$ GABA_A receptor function by zuranolone + other GABA_A receptor modulators

Treatment group	EC ₅₀ (nmol/L)	E _{max} (%)	CTD
Zuranolone + Diazepam	199	262	4.2.1.4-02
Zuranolone + Pentobarbital	605	1055	4.2.1.4-03
Zuranolone + Propofol (3 μ mol/L)	1385	636	4.2.1.4-05

3.4.2 Effects on direct gating of GABA_A receptors (Reference data CTD 4.2.1.4-06)

Using Ltk cells expressing human $\alpha_1\beta_2\gamma_2$ GABA_A receptors, the effects of zuranolone (10 μ mol/L) in combination with diazepam (0.1 μ mol/L) or pentobarbital (300 μ mol/L) on the direct gating activity of GABA_A receptors were examined in the absence of GABA using a whole-cell patch-clamp technique. Compared with zuranolone alone, coadministration of diazepam increased the current amplitude by an average of 34%. Compared with zuranolone alone, coadministration of pentobarbital increased the current amplitude by an average of 120%.

3.4.3 Effects in seizure models

In a mouse model of pentylenetetrazol (a GABA receptor antagonist)-induced seizures, the effects of zuranolone (0.1-1 mg/kg) coadministered intraperitoneally with diazepam (0.1 mg/kg) on seizures were assessed for drug interaction evaluations. Zuranolone 0.1 mg/kg alone or diazepam 0.1 mg/kg alone did not alter the latency to tonic seizures, compared with vehicle. Coadministration of zuranolone 0.1 mg/kg with diazepam 0.1 mg/kg increased the latency to tonic seizures, compared with vehicle (Reference data CTD 4.2.1.4-07).

In a rat model of lithium-pilocarpine-induced status epilepticus, the effects of zuranolone (0.3-3 mg/kg) coadministered intravenously with diazepam (10 mg/kg) at 60 minutes after the onset of seizures on the EEG power spectrum were assessed for drug interaction evaluations. No reduction in the EEG power was observed after administration of diazepam alone or zuranolone 0.3 to 1 mg/kg alone. Zuranolone 1 mg/kg in combination with diazepam 10 mg/kg resulted in a reduction in the EEG power in multiple frequency bands (Reference data CTD 4.2.1.4-08).

3.R Outline of the review conducted by PMDA

3.R.1 Primary pharmacodynamic studies

The applicant's explanation about the effects of zuranolone on depressive symptoms:

The pathogenesis of depression is not fully understood, and the monoamine hypothesis, focusing on the deficiencies of neurotransmitters such as serotonin, noradrenaline, and dopamine in the brain, has mainly been proposed to date. On the other hand, as shown below, decreases in the levels of an inhibitory neurotransmitter, GABA and a neuroactive steroid that enhances GABA_A receptor function, allopregnanolone (*Nat Rev Neurosci.* 2005; 6: 565-75) and associated decreased GABA signaling in patients with depression and animal models of depression have been reported. Neuronal hyperexcitability induced by decreased GABA signaling results in dysregulated network activity of excitatory and inhibitory neurons, which

is also considered to play a role in the development of depression.

- GABA and allopregnanolone levels in the brain regions related to emotion regulation and stress response, such as the prefrontal cortex, are lower in depressed patients compared with healthy adult individuals (*Hum Brain Mapp.* 2016; 37: 3337-52, *Psychopharmacology (Berl)*. 2014; 231: 3569-80), and a negative correlation exists between GABA and allopregnanolone levels and the severity of depression (*Proc Natl Acad Sci USA*. 1998; 95: 3239-44, *J Psychiatr Res.* 1988; 22: 159-64).
- In animal models of depression, lowered expressions of the genes and proteins related to GABA synthesis and a decrease in spontaneous inhibitory postsynaptic currents in the prefrontal cortex (*Transl Psychiatry*. 2016; 6: e910), a decrease in allopregnanolone levels in the prefrontal cortex, hippocampus, and amygdala (*Neurobiol Stress*. 2020; 12: 100218, *Biol Psychiatry*. 2023; 94: 249-61), and decreases in phasic and tonic GABA currents mediated by γ subunit- and δ subunit-containing GABA_A receptors, respectively (*Mol Brain*. 2014; 7: 32, *Transl Psychiatry*. 2023; 13: 229, etc.) were observed. In mice lacking γ subunit or δ subunit, decreases in phasic and tonic GABA currents and anxiety and depressive-like behaviors were observed (*Biol Psychiatry*. 2023; 94: 249-61, *Front Cell Neurosci*. 2019; 13: 87).

GABA_A receptors are pentameric Cl⁻ channels comprised of 2 α , 2 β , and 1 auxiliary subunits (typically, γ or δ). GABA_A receptors containing the γ subunit are the predominant subtype present at post-synaptic sites where high concentrations of GABA are present and play a role in phasic inhibition of neuronal excitability by generating phasic GABA currents. GABA_A receptors containing the δ subunit are present largely extrasynaptically, where low concentrations of GABA are present and play a role in tonic inhibition of neuronal excitability by generating tonic GABA currents (*Transl Psychiatry*. 2023; 13: 228, *Nat Rev Neurosci*. 2005; 6: 215-29, etc.). Given the following results from non-clinical studies of zuranolone etc., an allopregnanolone analog, zuranolone is expected to exert antidepressant effects by potentiating phasic and tonic GABA currents mediated by γ subunit-containing and δ subunit-containing GABA_A receptors, respectively. Enhancement of δ subunit-containing GABA_A receptor function should be more important for the exertion of antidepressant effects of zuranolone.

- While the currently approved benzodiazepines bind at a cleft between α and γ subunits of the GABA_A receptor (*Curr Top Med Chem*. 2002; 2: 833-9), a neuroactive steroid, allopregnanolone and its analog, zuranolone bind to the α and β subunits of the GABA_A receptor (*Pharmacol Ther*. 2007; 116: 7-19, *Neuropharmacology*. 2009; 56: 149-154, etc.) and therefore act on δ subunit-containing GABA_A receptors as well as γ subunit-containing GABA_A receptors [see Sections 3.1.1.2 and 3.1.1.3].
- Zuranolone potentiated GABA-evoked currents in cells expressing human GABA_A receptors containing the γ subunit or δ subunit [see Sections 3.1.1.2 and 3.1.1.3]. Zuranolone enhanced phasic and tonic GABA currents in mouse and rat brain slices [see Sections 3.1.1.5 and 3.1.1.6].
- An increase in the β -band EEG power is known as a biomarker of GABA_A receptor PAM activity (*J Pharmacol Exp Ther*. 2002; 302: 1158-67, *J Pharmacol Exp Ther*. 2003; 304: 88-101, etc.), and the θ -band EEG power was potentiated by neuroactive steroids such as allopregnanolone, which enhance δ subunit-containing GABA_A receptor function (*Biol Psychiatry*. 2022; 91: 283-93). Zuranolone elevated the power of both the β and θ frequency bands [see Section 3.1.2.2].

- In a mouse model of depression induced by social defeat stress, zuranolone increased the interaction time with other mice [see Section 3.1.2.1].
- After administration of a neuroactive steroid such as allopregnanolone, i.e., a GABA_A receptor PAM, in mice with depression induced by social defeat stress, the interaction time with other mice and the basolateral amygdala θ power increased, and these effects were attenuated in extrasynaptic GABA_A receptor δ -subunit knockout mice. Meanwhile, the interaction time with other mice or the basolateral amygdala θ power did not increase even after administration of an approved benzodiazepine, diazepam, in mice with depression induced by social defeat stress (*Front Cell Neurosci.* 2024; 17: 1274459).

PMDA's conclusion:

Based on the submitted study results etc., zuranolone is expected to exert antidepressant effects by binding to δ subunit-containing GABA_A receptors as well as γ subunit-containing GABA_A receptors and enhancing GABA_A receptor function.

3.R.2 Safety pharmacology studies

The applicant's explanation about the findings observed in safety pharmacology studies:

With respect to the effects of zuranolone on the central nervous and respiratory systems, clinical signs and changes in functional observational battery (FOB) indicative of severe sedation were observed in a CNS study in rats and a cardiovascular study in dogs, and a decrease in respiratory rate was observed in a respiratory study in rats. Also in repeated-dose toxicity studies in rats and dogs, decreased body temperature and abnormal breathing were noted at doses associated with severe sedation [see Section 5.2]. Given that zuranolone binds to GABA_A receptors and enhances GABA_A receptor function [see Section 3.R.1], sedation, decreased body temperature, and abnormal breathing observed in non-clinical studies are considered consistent with the exaggerated pharmacological effects of zuranolone. The no-observed-effect level (NOEL) was not identified in the CNS study, and zuranolone exposures (the $C_{\max}$ values of 102 and 40.6 ng/mL, respectively)⁹⁾ at the doses at which no sedation-related findings were observed in the cardiovascular study (dog) and the respiratory study (rat) (0.4 mg/kg and 3 mg/kg, respectively) are close to the human exposure ($C_{\max} = 67.2$ ng/mL)¹⁰⁾ at the clinical dose of 30 mg/day. In addition, when zuranolone exposure ($C_{\max}$) at which severe sedation was observed in the repeated-dose toxicity studies was compared with the human exposure ($C_{\max} = 67.2$ ng/mL) at the clinical dose of 30 mg/day, the dog to human exposure ratio was 4.5 to 10.9, and the rat to human exposure ratio was 6.1 to 13.7. However, Japanese and foreign clinical studies of zuranolone raised no concerns about sedation and respiratory effects. Thus, though the CNS and respiratory effects of zuranolone still deserve attention after marketing, clinically relevant safety concerns are unlikely to arise.

With respect to the cardiovascular effects of zuranolone, decreases in systolic blood pressure and mean blood pressure and increased heart rate observed in dogs were considered exaggerated pharmacological effects of

9) Since no zuranolone exposure data were collected, zuranolone exposure on Day 1 in the zuranolone 0.4 mg/kg group in a 2-week repeated-dose toxicity study in dogs and zuranolone exposure on Day 1 in the zuranolone 3 mg/kg group in a 2-week repeated-dose toxicity study in rats were used.

10) The median $C_{\max}$ following repeated oral administration of zuranolone 30 mg/day predicted from the PPK model [see Section 6.2.6]

zuranolone, i.e., enhanced GABA_A receptor function, based on the following points. Changes in ECG parameters observed in dogs were considered secondary to increased heart rate.

- Administration of GABA_A receptor PAM, anesthetics etc., in dogs resulted in decreased blood pressure and a transient increase in heart rate (*Vet Anaesth Analg.* 2008; 35: 451-62, *J Small Anim Pract.* 1987; 28: 623-637, etc.).
- Clinical signs indicative of severe sedation were observed at doses (≥ 1 mg/kg) associated with cardiovascular effects in the cardiovascular study in dogs (Table 8).
- In the cardiovascular study in dogs, increased heart rate was temporally associated with decreased blood pressure.

Although zuranolone exposure ($C_{\max} = 102$ ng/mL) at the NOEL (0.4 mg/kg) in the cardiovascular study in dogs is close to the human exposure ($C_{\max} = 67.2$ ng/mL) at the clinical dose of 30 mg/day, given that there were no decreased blood pressure, increased heart rate, or abnormalities in ECG parameters in Japanese and foreign clinical studies of zuranolone including the QT/ corrected QT (QTc) evaluation study [see Sections 6.2.5.1 and 7.R.3], etc., zuranolone in clinical use is unlikely to affect the cardiovascular system.

PMDA's conclusion on the findings observed in safety pharmacology studies:

Given the applicant's explanation about non-clinical and clinical studies of zuranolone, zuranolone has the potential to affect the central nervous and respiratory systems in humans. Thus, a review of the safety of zuranolone in humans will be continued in Sections 7.R.3.2 and 7.R.3.3.

4. Non-clinical Pharmacokinetics and Outline of the Review Conducted by PMDA

The applicant submitted the results from absorption, distribution, metabolism, and excretion studies in mice, rats, dogs, and rabbits as the non-clinical pharmacokinetic studies of zuranolone. Zuranolone concentrations in biomaterials were determined using liquid chromatography-tandem mass spectrometry (LC-MS/MS) (lower limit of quantification [LLOQ], 1.00 or 5.00 ng/mL). In studies using ¹⁴C-zuranolone, radioactivity concentrations in biomaterials were determined using liquid scintillation counter, and tissue radioactivity concentrations were determined using quantitative whole-body autoradiography. The main study results are described below.

4.1 Absorption

4.1.1 Single-dose studies

Table 11 shows the plasma pharmacokinetic parameters of zuranolone following a single intravenous or oral dose of zuranolone in mice, rats, and dogs under non-fasting conditions.

Table 11. Plasma pharmacokinetic parameters of zuranolone following a single intravenous or oral dose of zuranolone

Species	Route of administration	Dose (mg/kg)	Sex No. of animals	C _{max} (ng/mL)	t _{max} (h) ^{a)}	t _{1/2} (h)	AUC (ng·h/mL) ^{b)}	Bioavailability (%) ^{c)}	CTD
Mouse ^{d)}	IV	5	2M/time point	3529	0.083	0.876	3589		Reference data 4.2.2.2-01
	Oral	10	2M/time point	1646	1.00	1.64	4435	61.8	
Rat	IV	1	3M	538 ± 106	0.083 (0.083, 0.083)	1.15 ± 0.170	538 ± 77.6		Reference data 4.2.2.2-10
	Oral	3	3M	305 ± 23.8	1.00 (1.00, 1.00)	1.88 ± 0.0334	853 ± 83.2	52.9 ± 5.16	
		5	3M	345 ± 97.0	1.00 (0.50, 1.50)	8.77 ± 6.56	1927 ± 729	58.3 ± 21.6	
Dog ^{e)}	IV	1	2M	1422, 1429	0.083, 0.167	11.2, 12.5	6150, 6948		Reference data 4.2.2.2-18
	Oral	3.72	2M	1309, 1643	1.00, 1.50	10.7, 15.0	13304, 20088	77.7, 58.1	

Mean or Mean ± SD

a) Median or Median (Min., Max.)

b) AUC_{0-∞} in mice and rats, AUC_{0-last} in dogs

c) (Dose-normalized AUC after oral administration/dose-normalized AUC after intravenous administration) × 100

d) Parameters were calculated based on the mean plasma concentration at each time point.

e) Individual values

4.1.2 Repeated-dose studies

Toxicokinetics were evaluated in repeated oral dose toxicity studies of zuranolone in rats, dogs, and rabbits. Table 12 shows the plasma pharmacokinetic parameters of zuranolone. There were sex-related differences in the pharmacokinetics of zuranolone in rats. The applicant explained that this was due to the reported sex differences in the expression levels of cytochromes P450 isoforms and sex-specific isoforms of cytochromes P450 in rats (*J Vet Med Sci.* 2010; 72: 471-9, *Drug Metab Rev.* 1998; 30: 441-98).

Table 12. Plasma pharmacokinetic parameters of zuranolone following repeated oral administration of zuranolone

Species	Sex	Doses (mg/kg/day)	Sampling time point	Number of animals	C _{max} (ng/mL)	t _{max} (h) ^{a)}	AUC _{0-last} (ng·h/mL)	CTD
Rat ^{b)}	Male	3	Day 1	3/time point	295	8	929	4.2.3.2-09
			Day 91	3/time point	214	2	1760	
			Day 182	3/time point	168	4	1690	
		10	Day 1	3/time point	313	2	3760	
			Day 91	3/time point	485	2	6150	
			Day 182	3/time point	350	6	4560	
		30	Day 1	3/time point	562	6	8020	
			Day 91	3/time point	513	2	6560	
			Day 182	3/time point	429	4	6350	
	Female	0.5	Day 1	3/time point	80.3	2	845	
			Day 91	3/time point	126	2	1280	
			Day 182	3/time point	83.9	2	1310	
		1.5	Day 1	3/time point	241	2	2600	
			Day 91	3/time point	263	1	4010	
			Day 182	3/time point	295	2	4060	
		5	Day 1	3/time point	560	2	7690	
			Day 91	3/time point	721	4	8680	
			Day 182	3/time point	564	2	7590	
Dog	Male	0.6	Day 1	4	157 ± 14.9	3 (2, 6)	1990 ± 365	4.2.3.2-15
			Day 37	4	173 ± 2.2	2 (2, 4)	1870 ± 229	
			Day 140	4	184 ± 32.1	2 (2, 2)	2500 ± 343	
			Day 273	4	183 ± 14	2 (1, 4)	2500 ± 280	
		1.2	Day 1	7	354 ± 73.7	2 (1, 4)	5000 ± 1050 ^{c)}	
			Day 37	7	372 ± 72.4	4 (2, 8)	5640 ± 838	
			Day 140	7	448 ± 75.8	4 (2, 6)	7550 ± 1230	
			Day 273	7	467 ± 75.8	4 (2, 8)	7500 ± 1350	
		2.5	Day 1	7	474 ± 83.5	4 (1, 8)	7630 ± 1870	
			Day 37	7	796 ± 247	1 (1, 6)	10900 ± 2810	
			Day 140	7	792 ± 270	4 (2, 6)	12200 ± 3920	
			Day 273	6	621 ± 53.6	4 (2, 6)	9840 ± 837	
	Female	0.6	Day 1	4	146 ± 8.54	2 (1, 2)	1750 ± 209	
			Day 37	4	155 ± 16.6	1.5 (1, 2)	1930 ± 243	
			Day 140	4	171 ± 44.4	2 (2, 4)	2240 ± 440	
			Day 273	4	196 ± 25.9	2 (2, 2)	2480 ± 430	
		1.2	Day 1	7	265 ± 40.4	4 (2, 8)	3500 ± 422	
			Day 37	7	291 ± 60.2	2 (1, 8)	4010 ± 829	
			Day 140	7	356 ± 73.3	2 (1, 4)	4910 ± 1030	
			Day 273	7	352 ± 64.6	2 (1, 4)	5300 ± 971	
		2.5	Day 1	7	364 ± 108	2 (2, 6)	5620 ± 1610	
			Day 37	6	604 ± 120	4 (2, 6)	9290 ± 1510	
			Day 140	6	709 ± 128	2 (1, 2)	11500 ± 2190	
			Day 273	6	730 ± 148	2 (2, 4)	11500 ± 2780	
Rabbit	Female	25	Day 1	1 ^{d)}	21.1	6	256	4.2.3.5-06
			Day 13	1 ^{d)}	20.9	4	291	
		125	Day 1	4	19.8 ± 20.3	5 (1, 8)	395 ± 423	
			Day 13	3 ^{e)}	39.1 ± 32.4	2 (1, 6)	663 ± 563	
		500	Day 1	4	16.9 ± 16.6	16 (6, 24)	233 ± 301	
			Day 13	4	21.1 ± 23.9	5 (1, 8)	260 ± 401	

Mean or Mean ± SD

a) Median or Median (Min., Max.)

b) Parameters were calculated based on the mean plasma concentration at each time point.

c) n = 6

d) Toxicokinetics were evaluated in 4 animals. Since plasma concentrations were below the LLOQ at many time points in 3 of the 4 animals, pharmacokinetic parameters (individual values) in 1 animal are presented.

e) Toxicokinetics were evaluated in 4 animals. Since plasma concentrations were below the LLOQ at all time points in 1 of the 4 animals, pharmacokinetic parameters were calculated based on the data from the 3 animals.

4.2 Distribution

4.2.1 Tissue distribution

Following a single oral dose of 10 mg/kg of ¹⁴C-zuranolone in male albino rats and a single oral dose of 1.5 mg/kg of ¹⁴C-zuranolone in female albino rats (1/sex/time point), radioactivity concentrations in different

tissues¹¹⁾ were determined up to 840 hours post-dose. The highest levels of radioactivity were found in most tissues examined¹²⁾ by 8 hours post-dose. In males, the highest level of radioactivity was found in the whole blood at 4 hours post-dose. At 4 hours post-dose, radioactivity concentrations in the tissues other than the adrenal gland, bone, cerebellum, cerebrum, medulla oblongata, olfactory bundle, cecum, lens, eyes, uveal tract, preputial gland, skin, and spinal cord were higher than that in the whole blood. In females, the highest level of radioactivity was found in the whole blood at 8 hours post-dose. At 8 hours post-dose, radioactivity concentrations in the tissues other than the bone, lens, and eyes¹³⁾ were higher than that in the whole blood. By 168 hours post-dose, radioactivity was undetectable in the tissues other than the liver in male rats and in the tissues other than the adrenal gland, fat (abdominal), brown fat, large intestine, liver, ovary, preputial gland, and skin in female rats.

Following a single oral dose of 10 mg/kg of ¹⁴C-zuranolone in male pigmented rats (1/time point), radioactivity concentrations in different tissues¹⁴⁾ were determined up to 840 hours post-dose. There was no trend towards major differences in tissue distribution between pigmented and albino rats. Radioactivity was undetectable in the tissues other than the liver at 168 hours post-dose, and there was no specific distribution of radioactivity to melanin-containing tissues (pigmented skin and uveal tract) (CTD 4.2.2.2-16).

Following oral administration of 10 mg/kg of zuranolone in male mice (3/time point), the brain distribution of zuranolone was determined. The highest levels of zuranolone were found in the plasma and brain at 0.5 hours post-dose, and the brain to plasma concentration ratio was 1.56. The brain to plasma zuranolone exposure (AUC_{0-last}) ratio was 1.40 (Reference data CTD 4.2.2.2-02).

Following oral administration of 5 mg/kg of zuranolone in male rats (2/time point), the brain distribution of zuranolone was determined. The highest levels of zuranolone were found in the plasma and brain at 1 hour post-dose, and the brain to plasma concentration ratio was 3.48. The brain to plasma zuranolone exposure (AUC_{0-last}) ratio was 3.22 (Reference data CTD 4.2.2.2-10).

4.2.2 Protein binding and distribution in blood cells

When zuranolone (10 µmol/L) was added to the plasma from mouse, rat, and dog, the plasma protein binding of zuranolone as determined by an equilibrium dialysis method was 99.0%, 98.8%, and 98.4%, respectively (Reference data CTD 5.3.2.1-03).

11) adrenal gland, arterial walls, bile, blood, bone, bone marrow, cerebellum, cerebrum, medulla oblongata, olfactory bundle, bulbourethral gland (males only), cecum, esophageal contents, stomach contents, diaphragm, epididymis (males only), esophagus, exorbital lacrimal gland, lens, uveal tract, eyes, fat (abdominal), fat (brown), Harderian gland, intra-orbital lachrymal gland, kidney cortex, kidney medulla, kidney, large intestine, liver, lung, lymph node, muscle, myocardium, nasal turbinates, ovary (females only), pancreas, pituitary gland, preputial gland, prostate gland (males only), salivary gland, seminal vesicle (males only), skin, small intestine, spinal cord, spleen, stomach, stomach mucosa, stomach wall, testis (males only), thymus, thyroid, urinary bladder, urine, uterus (females only)

12) Tissues other than the large intestine in males and tissues other than the lens in females

13) Radioactivity in the intra-orbital lachrymal gland was not measured at 8 hours post-dose in females.

14) adrenal gland, arterial walls, bile, blood, bone, bone marrow, cerebellum, cerebrum, medulla oblongata, olfactory bundle, bulbourethral gland, cecum, esophageal contents, stomach contents, diaphragm, epididymis, esophagus, exorbital lacrimal gland, lens, uveal tract, eyes, fat (abdominal), fat (brown), Harderian gland, intra-orbital lachrymal gland, kidney cortex, kidney medulla, kidney, large intestine, liver, lung, lymph node, muscle, myocardium, nasal turbinates, pancreas, pituitary gland, preputial gland, prostate gland, salivary gland, seminal vesicle, skin (non-pigmented), skin (pigmented), small intestine, spinal cord, spleen, stomach, stomach mucosa, stomach wall, testis, thymus, thyroid, urinary bladder, urine

Following a single oral dose of 10 mg/kg of ^{14}C -zuranolone in male albino rats and a single oral dose of 1.5 mg/kg of ^{14}C -zuranolone in female albino rats (3/sex/time point), the blood to plasma radioactivity concentration ratios were <1 (0.397-0.880) up to 4 hours post-dose and 1.56 to 2.28 at 8 to 24 hours post-dose in males and <1 (0.325-0.889) up to 8 hours post-dose, 1.32 at 12 hours post-dose, and 0.826 at 24 hours post-dose in females (CTD 4.2.2.2-16).

4.2.3 Placental transfer

Placental transfer of zuranolone has not been studied, and no data on zuranolone fetal transfer have been obtained. However, given that zuranolone has been suggested to be highly lipid-soluble, and that fetal malformations etc. (direct effects of zuranolone could not be ruled out) were observed in an embryo-fetal development study in rats (CTD 4.2.3.5-04) [see Section 5.R.1], zuranolone may cross the placenta into the fetus.

4.3 Metabolism

4.3.1 *In vitro* metabolism

When zuranolone 100 $\mu\text{mol/L}$ was added to rat or dog hepatocytes and incubated at 37°C for 240 minutes, 4 mono-hydroxylated metabolites, M1 to 4, were detected in hepatocytes of both species (Reference data CTD 5.3.2.2-08).

4.3.2 *In vivo* metabolism

Rats (3/sex/time point) received a single oral dose of ^{14}C -zuranolone 10 mg/kg (males) or 1.5 mg/kg (females). In the plasma collected up to 48 hours post-dose, unchanged zuranolone (35.1% of the total radioactivity in plasma) and M76 (dihydroxy-zuranolone) (14.0% of the total radioactivity in plasma) were detected in males, and unchanged zuranolone (61.5% of the total radioactivity in plasma) was detected in females. Other metabolites detected accounted for <10% of the total radioactivity in plasma in both males and females.

Intact and bile duct cannulated rats (3/sex each) received a single oral dose of ^{14}C -zuranolone 10 mg/kg (males) or 1.5 mg/kg (females), and its metabolites in urine, bile (bile duct cannulated rats only), and feces (intact rats only) were identified. In the urine collected up to 48 hours post-dose, unchanged zuranolone was not detected in intact or bile duct cannulated male or female rats, and many metabolites accounting for <2% of the administered radioactivity were present. In the bile collected up to 48 hours post-dose in males, unchanged zuranolone was not detected; M53 (structurally unidentified) (10.2% of the total administered radioactivity) was the most abundant metabolite; and many metabolites were present. In the bile collected up to 72 hours post-dose in females, unchanged zuranolone (1.6% of the total administered radioactivity) was detected; M115 (structurally unidentified) (20.4% of the total administered radioactivity) was the most abundant metabolite; and many metabolites were present. In the feces collected up to 72 hours post-dose in males, unchanged zuranolone was not detected; M108 (hydroxy-zuranolone sulfate) (6.3% of the total administered radioactivity) was the most abundant metabolite; and many metabolites were present. In the feces collected up to 72 hours post-dose in females, unchanged zuranolone (4.8% of the total administered radioactivity) was detected; M108 (7.4% of the total administered radioactivity) was the most abundant metabolite; and many metabolites were

present (CTD 4.2.2.2-16).

Dogs (3/sex) received a single oral dose of ^{14}C -zuranolone 1 mg/kg. In the plasma collected up to 72 hours post-dose, unchanged zuranolone (55.3%-67.6% of the total radioactivity in plasma) and M20 (hydroxy-zuranolone) (8.7%-9.7% of the total radioactivity in plasma) were detected, and other metabolites detected accounted for <5% of the total radioactivity in plasma. In the urine collected up to 120 hours post-dose, unchanged zuranolone (4.2%-6.4% of the total administered radioactivity) and M13 (hydroxy-oxopyrazole-zuranolone) (2.7%-2.8% of the total administered radioactivity) were detected, and other metabolites detected accounted for <1% of the total administered radioactivity. In the feces collected up to 96 hours post-dose, unchanged zuranolone (14.8%-19.0% of the total administered radioactivity), M13 (7.0%-8.0% of the total administered radioactivity), M20 (4.1%-4.9% of the total administered radioactivity), and M26 (oxopyrazole-zuranolone) (3.2%-5.3% of the administered radioactivity) were detected, and other metabolites detected accounted for <2% of the total administered radioactivity (CTD 4.2.2.2-20).

4.4 Excretion

4.4.1 Urinary, fecal, and biliary excretion

Following a single oral dose of 10 mg/kg of ^{14}C -zuranolone in intact male rats and bile duct cannulated male rats and a single oral dose of 1.5 mg/kg of ^{14}C -zuranolone in intact female rats and bile duct cannulated female rats (3/sex each), the cumulative recoveries of the administered radioactivity in urine and feces over 168 hours were 9.1% to 15.2% and 76.9% to 85.7%, respectively, in intact rats and the cumulative recoveries of the administered radioactivity in urine, feces, and bile over 168 hours were 4.2% to 10.8%, 2.2% to 3.5%, and 81.6% to 83.0%, respectively, in bile duct cannulated rats (CTD 4.2.2.2-16).

Following a single oral dose of 1 mg/kg of ^{14}C -zuranolone in dogs (3/sex), the cumulative recoveries of the administered radioactivity in urine and feces over 168 hours were 12.2% to 13.0% and 54.9% to 68.3%, respectively (CTD 4.2.2.2-20).

4.4.2 Excretion into milk

Zuranolone has been shown to be excreted in human milk [see Section 6.2.2.3].

4.R Outline of the review conducted by PMDA

PMDA concluded that the submitted data gave a grasp of the *in vivo* behavior of zuranolone to a certain extent, and that its non-clinical pharmacokinetic profile has been characterized.

5. Toxicology and Outline of the Review Conducted by PMDA

The applicant submitted the results from the following toxicity studies of zuranolone: single-dose toxicity, repeated-dose toxicity, genotoxicity, carcinogenicity, and reproductive and developmental toxicity studies and other studies (juvenile animal and dependence studies). The main study results are described below. Unless otherwise specified, 2% (w/v) polyvinylpyrrolidone-vinyl acetate (PVP-VA) in deionized (DI) water was used as vehicle in *in vivo* studies.

5.1 Single-dose toxicity

Single oral dose toxicity studies of zuranolone were conducted in rats and dogs, and the approximate lethal doses by oral route were determined to be 5 mg/kg in rats and >3 mg/kg in dogs (Table 13).

Table 13. Overview of single oral dose toxicity studies

Test system	Route of administration	Doses (mg/kg/day)	Noteworthy findings	Approximate lethal dose (mg/kg)	CTD
Male and female rats (SD)	Oral gavage	Males: 20, 25, 30, 35, 40 Females: 2.5, 4, 5, 7.5, 10	Mortalities ^{a)} : 10 (2 of 3 females), 5 (3 of 3 females) ≥2.5: hypoactivity, hypersensitivity/reactivity to touch and/or sound, myoclonus, tremors, exophthalmos, loss of body tone	Males: >40 Females: 5	Reference data 4.2.3.1-01
Male and female dogs (Beagle)	Oral gavage	1, 1.5, 2, 2.5, 3	≥1: sedation, impaired equilibrium, hypoactivity, tremors, partial or complete closure of both eyes, hypersensitivity to touch and/or sound, loss of body tone ≥2.5: severe sedation	>3	Reference data 4.2.3.1-03
Male dog (Beagle)	Oral gavage ^{b)}	3.5 ^{c)}	severe sedation, ataxia, hypoactivity, lateral or sternal recumbency, partially closed eyes, closed eyes, loss of consciousness, twitching, tremors, paddling, increase/decrease in muscle tone, non-sustained convulsions, decrease/increase in respiratory rate, reduced body temperature	>3.5	Reference data 4.2.3.2-10

a) These females were euthanized moribund due to prolonged duration of severe sedation and body cool to touch.

b) Vehicle: 15% (w/v) sulfobutylether-β-cyclodextrin

c) The study was conducted as a 7-day repeated-dose toxicity study, and the acute toxicity of zuranolone was assessed in the study. The study was terminated early on Day 3 because some animals were euthanized moribund due to severe convulsions and tachycardia.

5.2 Repeated-dose toxicity

Repeated oral dose toxicity studies were conducted in rats (2, 13, and 26 weeks) and dogs (2, 13, 29, 39, and 44 weeks) (Table 14).

The principal finding observed in rats and dogs was sedation, which was considered related to the pharmacologic effects of zuranolone. The findings of sedation accompanied by severe clinical signs including mortalities and moribund sacrifices were considered zuranolone-related toxicological findings.

Zuranolone exposures (AUC_{0-last}) at the no-observed-adverse effect levels (NOAELs) in the repeated oral dose toxicity studies in rats (26 weeks) and dogs (39 weeks) (10 mg/kg/day in male rats, 5 mg/kg/day in female rats, 1.2 mg/kg/day in dogs) were 4.8-fold (male rats), 8.0-fold (female rats), and 6.8-fold (dogs) the human exposure ($AUC_{0-24h} = 943.7 \text{ ng}\cdot\text{h/mL}$)¹⁵⁾ following oral administration of the clinical dose (30 mg/day) of zuranolone.

15) The median AUC_{0-24h} following repeated oral administration of 30 mg/day of zuranolone predicted from the PPK model [see Section 6.2.6].

Table 14. Overview of repeated oral dose toxicity studies

Test system	Route of administration	Duration of dosing	Doses (mg/kg/day)	Noteworthy findings	NOAEL (mg/kg/day)	CTD
Male and female rats (SD)	Oral gavage	2 weeks (QD) + 2-week recovery period	Males: 0, ^{a)} 3, 10, 22.5 Females: 0, ^{a)} 1, 3, 8	Males 22.5: sedation, increased APTT, increased AST, increased urine pH, hepatocellular vacuolation Females 8: sedation, increased APTT, increases in AST/ALT, increased urine pH, increased liver weights, hepatocellular vacuolation These findings were reversible (male and female).	Males: 22.5 ^{b)} Females: 3 ^{b)} c)	4.2.3.2-06 Reference data 4.2.3.2-07
Male and female rats (SD)	Oral gavage	13 weeks (QD) + 4-week recovery period ^{d)}	Males: 0, 0, ^{e)} 0.8, 2.5, 8, 30 Females: 0, 0, ^{e)} 0.15, 0.5, 1.5, 5	Males 30: sedation, tremors, impaired equilibrium, hyper-reactivity to touch Females Mortality: 5 (1 of 15 females) 5 ^{f)} : sedation, tremors, impaired equilibrium, hyper-reactivity to touch These findings were reversible (male and female).	Males: 30 ^{f)} Females: 1.5	4.2.3.2-08
Male and female rats (SD)	Oral gavage	26 weeks (QD) + 4-week recovery period ^{g)}	Males: 0, 3, 10, 30 Females: 0, 0.5, 1.5, 5	Males 30: sedation, decreased activity, uncoordinated, lack of/impaired righting reflex, low carriage Females 5: sedation, decreased activity, uncoordinated, lack of/impaired righting reflex, low carriage These findings were reversible (male and female).	Males: 10 Females: 5 ^{f)}	4.2.3.2-09
Male and female dogs (Beagle)	Oral gavage	2 weeks (QD) + 2-week recovery period	0, ^{h)} 0.4, 1, 2.5	2.5: sedation, a decrease in core body temperature, increased heart rate These findings were reversible.	2.5 ^{f)}	4.2.3.2-13
Male and female dogs (Beagle)	Oral gavage	13 weeks (QD) + 4-week recovery period ^{h)}	0, 0, ^{e)} 0.06, 0.2, 0.6, 2	Mortality: 2 (1 of 6 males) transient collapse, legs stiffened, aggression upon handling, tonic convulsions 2: sedation, hypoactivity, impaired equilibrium, ataxia, vocalization, rigid muscle tone, tremors, increased heart rate These findings were reversible.	0.6	4.2.3.2-14
Male and female dogs (Beagle)	Oral gavage	39 weeks (QD) + 4-week recovery period ^{h)}	0, 0.6, 1.2, 2.5	Mortalities: 2.5 (2 of 7 females ^{k)}) tonic and clonic convulsions, unsteady on feet, circling, vocalization 2.5: sedation, hypoactivity, ataxia, impaired equilibrium, low carriage, tremors These findings were reversible.	1.2	4.2.3.2-15

Table 14. Overview of repeated oral dose toxicity studies (continued)

Male and female dogs (Beagle)	Oral gavage	2 weeks (QD) + 42-day non-dosing period or 21-day non-dosing period 6 cycles ¹⁾ each	0, 1.2, 2.5	Mortalities: 2.5 (1 of 6 males, 1 of 6 females) uncoordinated, decreased activity, lying on side, abnormal gait, tremors, panting, unable to support itself 2.5: salivation, convulsion, paddling, limb rigidity, twitches, vocalization, panting, lethargy, lack of pinch reflex These findings were reversible.	1.2	4.2.3.2-16
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a) Vehicle: PEG-8 caprylic/capric glycerides

b) The findings observed in males in the 22.5 mg/kg/day group and females in the 8 mg/kg/day group were all minimal or mild in severity, and histopathological examination revealed no degenerative changes such as necrosis and inflammation. Thus, these findings were considered non-adverse.

c) One of 6 females in the 8 mg/kg/day group for pharmacokinetic assessment died, which was considered related to severe sedation caused by zuranolone. Thus, the NOAEL was determined to be 3 mg/kg/day.

d) The 0 (vehicle), 0 (water), 1.5, 5, 8, and 30 mg/kg/day groups were included.

e) Water

f) The observed findings were associated with mild to moderate sedation related to pharmacological effects of zuranolone, and clinical pathology and histopathological examinations revealed no changes indicative of functional or structural impairment. Thus, these findings were considered non-adverse.

g) The 0, 1.5, 5, 10, and 30 mg/kg/day groups were included.

h) Vehicle: 30% HPβCD aqueous solution

i) The 0 (vehicle), 0 (water), 0.6, and 2 mg/kg/day groups were included.

j) The 0, 1.2, and 2.5 mg/kg/day groups were included.

k) One female was found dead during the recovery period, but its relationship to zuranolone could not be ruled out.

l) Taking account of the dosing regimen of zuranolone in humans, the study was conducted to assess the potential toxicity of zuranolone.

5.3 Genotoxicity

An *in vitro* bacterial reverse mutation assay, an *in vitro* chromosomal aberration assay in cultured mammalian cells, and an *in vivo* bone marrow micronucleus assay in rats were conducted. Zuranolone tested negative for genotoxicity (Table 15).

Table 15. Overview of genotoxicity studies

Type of study	Test system	Metabolic activation (Treatment time)	Concentrations or doses	Test result	CTD
<i>In vitro</i>	Bacterial reverse mutation assay	<i>Salmonella typhimurium</i> : TA98, TA100, TA1535, TA1537 <i>Escherichia coli</i> : WP2uvrA	S9+/+ 0, ^{a)} 15, 50, 150, 500, 1500, 5000 (µg/plate)	Negative	4.2.3.3-01
		PAPS and human liver S9	0, ^{a)} 1.5, 5.0, 15, 50, 150, 500, 1500, 5000 ^{b)} (µg/plate)	Negative	4.2.3.3-02
	Chromosomal aberration assay in cultured mammalian cells	CHO cells	S9+ (4 hours) 0, ^{a)} 5, 10, 15, 20, 25, 30, 35, 40, 45, 50 (µg/mL)	Negative	4.2.3.3-03
		CHO cells	S9- (4, 20 hours) 0, ^{a)} 1, 2.5, 5, 10, 15, 20, 25, 30, 35, 40, 50, 60, 70, 85, 100, 120 (µg/mL)		
<i>In vivo</i>	Rodent bone marrow micronucleus assay	Male rat (SD) bone marrow	0, ^{c)} 3, 10, 22.5, 30 (mg/kg, oral gavage, single dose)	Negative	4.2.3.3-06

a) Vehicle: DMSO

b) Test article precipitation was observed at ≥150 µg/plate, and the number of revertant colonies could not be counted at 5000 µg/plate due to precipitation.

c) Vehicle: PEG-8 caprylic/capric glycerides

5.4 Carcinogenicity

Carcinogenicity studies were conducted in Tg rasH2 mice and rats (Table 16). Zuranolone exposures (AUC_{0-24h}) at the NOELs for carcinogenicity in mice and rats (100 mg/kg/day in mice, 20 mg/kg/day in male rats, 1.5 mg/kg/day in female rats) were 5.4-fold (mice), 6.8-fold (male rats), and 4.8-fold (female rats) the human exposure (AUC_{0-24h} = 943.7 ng·h/mL)¹⁵⁾ following oral administration of the clinical dose (30 mg/day) of zuranolone.

Table 16. Overview of carcinogenicity studies

Table 16. Overview of carcinogenicity studies															
Test system	Route of administration	Duration of dosing	Major lesions		Doses (mg/kg/day)						NOEL for carcinogenicity (mg/kg/day)	CTD			
					0	0 ^{a)}		10	30	100					
Male and female mice (rasH2)	Oral gavage	26 weeks (QD)		N	25M25F	25M25F		25M25F	25M25F	25M25F		100	4.2.3.4-02		
Neoplastic lesions			None												
Non-neoplastic lesions			Hypertrophy of corpora lutea in the ovaries, lobular hyperplasia in the mammary gland (female)												
Male and female rats (SD)	Oral gavage	92 weeks (QD)			Doses (mg/kg/day)								Males: 20 Females: 1.5	4.2.3.4-01	
				Male					Female						
				0	0 ^{a)}	2	6	20	0	0 ^{a)}	0.2	0.6			1.5
			N	70	70	70	70	70	70	70	70	70			70
			Neoplastic lesions	None											
			Non-neoplastic lesions	None											

a) Water

5.5 Reproductive and developmental toxicity

A rat study of fertility and early embryonic development to implantation, embryo-fetal development studies in rats, mice, and rabbits, and a rat study for effects on pre- and postnatal development, including maternal function were conducted (Table 17). Zuranolone exposures (AUC_{0-last}) at the NOAELs for embryo-fetal development (7.5 mg/kg/day in rats, 100 mg/kg/day in mice, 125 mg/kg/day in rabbits) were 9.9-fold (rats), 5.8-fold (mice), and 0.7-fold (rabbits) the human exposure ($AUC_{0-24h} = 943.7 \text{ ng}\cdot\text{h/mL}$)¹⁵⁾ following oral administration of the clinical dose (30 mg/day) of zuranolone.

The applicant's explanation:

Since the frequency and severity of zuranolone-related sedation increased with increasing dose of zuranolone in an embryo-fetal development study in rabbits (CTD 4.2.3.5-06), the study was considered to be conducted at appropriate dose levels. However, as it was considered difficult to fully evaluate the effects of zuranolone on embryo-fetal development using the results from this study due to relatively large inter-individual differences in maternal plasma zuranolone concentrations (Table 12), an embryo-fetal developmental toxicity study was conducted in mice.

Table 17. Overview of reproductive and developmental toxicity studies

Type of study	Test system	Route of administration	Duration of dosing	Doses (mg/kg/day)	Noteworthy findings	NOAEL (mg/kg/day)	CTD
Fertility and early embryonic development to implantation	Male rat (SD)	Oral gavage	From 28 days prior to mating, through mating, and until 24 days after mating (QD)	0, 3, 10, 30	General toxicity: Mortalities: 30 (2 of 25 males) 30: sedation, hyperreactivity to touch, lack of righting reflex, cold to touch, rales and labored breathing Reproductive function: No noteworthy findings	Parental animals (General toxicity): 10 Parental animals (Reproductive function): 30	4.2.3.5-02
	Female rat (SD)	Oral gavage	From 15 days prior to mating through gestation day 7 (QD) Caesarean section: Gestation day 13	0, 1, 3, 10	General toxicity: Mortalities: 10 (2 of 24 females) 10: sedation, twitches, ungroomed coat, chromodacryorrhea, dehydration, ataxia, gasping, urine-stained abdominal fur Reproductive function: 10: persistent diestrus until Day 10, a reduction in the number of estrus stages	Parental animals (General toxicity): 3 Parental animals (Reproductive function): 10 ^{a)}	4.2.3.5-01

Embryo-fetal development	Female rat (SD)	Oral gavage	From gestation day 6 through gestation day 17 (QD) Caesarean section: Gestation day 21	0, 2.5, 7.5, 22.5	Dams: Mortalities: 22.5 (3 of 23 females) 22.5: convulsions, suspected dehydration, thin appearance, ungroomed/stained fur, abnormal sounds of breathing, labored breathing, mucoid/liquid vaginal discharge, reduced body weight gain, reduced food consumption ≥7.5: sedation, red discharge in the eye, twitches Embryos/fetuses: 22.5: decreases in gravid uterine/fetal weights, increases in postimplantation loss/early resorptions, exencephaly, herniated umbilicus, depressed eye bulge, absent/small lens, abnormalities in the thoracic vertebra/lumbar vertebra/rib/tail/caudal vertebra/sacral vertebra, a reduction in the average numbers of ossified bones of the hindlimbs ≥7.5: absent rib, fused rib, fused thoracic arch, absent thoracic vertebra	Parental animals (General toxicity): 2.5 Embryo-fetal development: 7.5 ^{b)}	4.2.3.5-04
	Female mouse (ICR)	Oral gavage	From gestation day 6 through gestation day 15 (QD) Caesarean section: Gestation day 18	0, 30, 100, 300	Dams: 300: reduced body weight gain, decreased gravid uterine weights Embryos/fetuses: 300: decreased fetal weights, cleft palate, increased incidence of incomplete ossification or unossification	Parental animals (General toxicity): 100 Embryo-fetal development: 100	4.2.3.5-12
	Female rabbit (NZW)	Oral gavage	From gestation day 7 through gestation day 19 (QD) Caesarean section: Gestation day 29	0, 25, 125, 500	Dams: Mortalities: 500 (2 of 25 females) reduced body weight, thin appearance, ungroomed fur 500: twitches, partly closed eye, labored breathing, abnormal sounds of breathing, decreased respiratory rate, lack of righting reflex, reduced body weight, decreased feces output ≥125: sedation, lying on side, reduced body weight gain, reduced food consumption Embryos/fetuses: 500: decreased fetal weights	Parental animals (General toxicity): 25 Embryo-fetal development: 125	4.2.3.5-06
Pre- and postnatal development, including maternal function	Female rat (SD)	Oral gavage	Dams: From gestation day 6 through lactation day 20 (QD)	0, 1, 4, 10	Dams: Mortalities: 10 (9 of 24 females ^{c)}), 4 (3 of 24 females) chromodacryorrhea, labored breathing, rales, excess salivation, gasping, convulsions, cold to touch, dehydration ≥4: sedation, tremors, reduced food consumption, hyperreactivity to touch F1: 10: cold to touch, reduced body weight ≥4: increased mortality at postnatal days 1-4, ^{d)} decreased pre-weaning body weights, reduced food consumption, decreased post-weaning body weights	Dams (General toxicity): 1 F1 development: 1	4.2.3.5-07

a) Effects on estrus cycles were observed during early dosing period only, there were no effects of zuranolone on copulation or fertility, and macroscopic examination at necropsy revealed no abnormal findings. Thus, the findings were considered non-adverse.

b) Since the incidences of the findings observed in the 7.5 mg/kg/day group fell within the historical control range of the testing facility, these findings were considered incidental.

c) In addition to 9 dams that died, 6 dams were euthanized due to no surviving pups by lactation day 4. Thus, all 8 surviving dams in the 10 mg/kg/day group and their pups were euthanized on lactation days 8-10.

d) Including pups that died presumably due to maternal cannibalism.

5.6 Other toxicity studies

5.6.1 Phototoxicity

Zuranolone does not absorb light within the range of natural sunlight (290-700 nm) and does not have a Molar Extinction Coefficient greater than 1000 L/mol/cm at any wavelength between 290 and 700 nm. Thus, the risk of phototoxicity of zuranolone was considered low.

5.6.2 Juvenile animal studies

Two toxicity studies in juvenile rats (7 or 22 days of age) were conducted (Table 18). Zuranolone exposures (AUC_{0-24h}) at the NOAELs in a repeated oral dose toxicity study in juvenile rats (10 mg/kg/day in males, 3→1 mg/kg/day in females¹⁶⁾) were 3.9-fold (juvenile male rats) and 2.7-fold (juvenile female rats) the human exposure ($AUC_{0-24h} = 943.7 \text{ ng}\cdot\text{h/mL}$)¹⁵⁾ following oral administration of the clinical dose (30 mg/day) of zuranolone.

Table 18. Overview of juvenile animal toxicity studies

Test system	Route of administration	Duration of dosing	Doses (mg/kg/day)	Noteworthy findings	NOAEL (mg/kg/day)	CTD
Male and female juvenile rats (SD) ^{a)}	Oral gavage	From postnatal day 22 through postnatal day 71 (QD) + 6-week recovery period	Males: 0, 3, 10, 30 Females ^{b)} : 0, 3→1, 10→3, 30→10	Males Mortalities: 30 (12 of 81 males) 30: sedation, tremors, shallow breathing, labored breathing, reduced food consumption, reduced body weight gain, increased activity Females Mortalities: 30 (24 of 81 females), 10 (1 of 81 females) 30 → 10: uncoordinated gait, tremors, shallow breathing, labored breathing, reduced food consumption, delayed age of vaginal patency ≥10→3: sedation These findings were reversible (male and female).	General toxicity: Males: 10 Females: 3→1 Reproductive function ^{c)} : Males: 10 Females: 10→3	4.2.3.5-09
Male and female juvenile rats (SD)	Oral gavage	Postnatal day 7 Single dose	0, 0, ^{d)} 2.5, 7.5	The potential for zuranolone to cause neurodegeneration in juvenile rats was assessed. ^{e)} Mortality: 7.5 (1 of 10 males) 7.5: sedation, hypoactivity, labored breathing, pale body, cool body, reduced body weight gain, increased neuronal disintegration within the subiculum	2.5 ^{f)}	4.2.3.5-10

a) Observation of sexual maturation (vaginal patency, balano-preputial separation), observation of estrous cycling, neurobehavioral assessments (motor activity, acoustic startle response, Morris water maze), and bone length and density measurements were performed.

b) The results from other toxicity studies in adult female rats showed that high-dose zuranolone may be less tolerable. Thus, the dose levels of zuranolone in female rats in this study were reduced from 3, 10, or 30 mg/kg/day to 1, 3, or 10 mg/kg/day, respectively, from postnatal day 36 onwards.

c) Fertility assessment in males in the 30 mg/kg/day group and females in the 30/10 mg/kg/day group was not performed due to high mortality.

d) Water

e) Using AmCuAg staining, disintegration of neuronal cell body and process due to degeneration/necrosis was assessed.

f) NOEL

5.6.3 Dependence

Drug discrimination, self-administration, and spontaneous withdrawal studies in rats were conducted (Table 19). Since zuranolone produced interoceptive effects similar to those of midazolam, which is also a GABA_A receptor PAM, and induced a withdrawal syndrome, zuranolone was considered to produce psychological and physical dependence.

16) The results from other toxicity studies in adult female rats showed that high-dose zuranolone may be less tolerable. Thus, the dose levels of zuranolone in female rats in this study were reduced from 3, 10, or 30 mg/kg/day to 1, 3, or 10 mg/kg/day, respectively, from postnatal day 36 onwards.

Table 19. Overview of dependence studies

Type of study	Test system	Test method	Noteworthy findings	CTD
Drug discrimination	Male rat (Lister Hooded)	Following intraperitoneal injection of zuranolone 0, ^{a)} 0.1, 0.3, 0.5, 1, or 3 mg/kg in rats that were trained to discriminate midazolam from vehicle (saline), drug discrimination and generalization were assessed.	Zuranolone at 0.5 and 1 mg/kg produced partial generalization to midazolam, with full generalization occurring at 3 mg/kg.	4.2.3.7-01
	Female rat (Lister Hooded)	Following intraperitoneal injection of zuranolone 0, ^{a)} 0.1, 0.3, 0.5, 1, or 2 mg/kg in rats that were trained to discriminate midazolam from vehicle (saline), drug discrimination and generalization were assessed.	Zuranolone at 0.5 and 1 mg/kg produced partial generalization to midazolam, with full generalization occurring at 2 mg/kg.	4.2.3.7-02
Self-administration	Female rat (Lister Hooded)	Rats that were trained to intravenously self-administer cocaine were free to intravenously self-administer zuranolone 0, ^{a)} 0.01, 0.03, or 0.05, midazolam, or cocaine 0.8 mg/kg, and the reinforcing properties of zuranolone were assessed.	Zuranolone did not maintain self-administration.	4.2.3.7-05
Spontaneous withdrawal	Male rat (Lister Hooded)	After repeated oral doses of zuranolone 0, 2, 6, or 20 mg/kg or CDP were administered for 28 days (QD), the drug was discontinued for 7 days, and withdrawal syndrome ^{b)} was assessed.	Discontinuation of zuranolone did not induce a withdrawal syndrome.	4.2.3.7-03
	Female rat (Lister Hooded)	After repeated oral doses of zuranolone 0, 0.5, 1.5, or 5 mg/kg or CDP were administered for 28 days (QD), the drug was discontinued for 7 days, and withdrawal syndrome ^{b)} was assessed.	Rats in the 5 mg/kg group spent significantly more time in the arena margins, showing the potential to induce a withdrawal syndrome.	4.2.3.7-04

a) Vehicle: 30% HPβCD aqueous solution

b) Withdrawal syndrome was assessed based on physiological parameters (body weight, food consumption, body temperature), locomotor activity, neurobehavioral findings, and clinical signs.

5.R Outline of the review conducted by PMDA

5.R.1 Use in pregnant women or women who may be pregnant

PMDA asked the applicant to explain the effects of zuranolone on embryo-fetal development observed in embryo-fetal developmental toxicity studies in rats and mice (Table 17) and then explain the appropriateness of the precautionary statement regarding use in pregnant women or women who may be pregnant.

The applicant's explanation:

The applicant discussed the embryo-fetal effects of zuranolone observed in the embryo-fetal developmental toxicity studies in rats and mice (Table 17) as follows:

- With respect to fetal malformations observed in the 22.5 mg/kg/day group of the embryo-fetal developmental toxicity study in rats (CTD 4.2.3.5-04), malformations occurred at various sites with low incidences, and the sites of malformations were not consistent. Given these findings etc., spontaneous findings in rats (*Congenit Anom (Kyoto)*. 2019; 59:125-31) may have manifested as zuranolone-related maternal toxicity. In fetuses in the 7.5 mg/kg/day group, absent rib, fused rib, fused thoracic arch, and absent thoracic vertebra were observed. The incidences of these findings in the 22.5 mg/kg/day group were within, or slightly above but generally comparable to, the historical control ranges of the testing facility or published literature values (*Congenit Anom (Kyoto)*. 2019; 59: 125-31). In the 7.5 mg/kg/day group, in which severe maternal toxicity was not observed, the incidences of all findings were within the historical control ranges. Based on the above etc., the NOAEL for embryo-fetal development in rats was determined to be 7.5 mg/kg/day.
- In the embryo-fetal developmental toxicity study in mice (CTD 4.2.3.5-12), cleft palates were observed in the 300 mg/kg/day group. Its incidence was within the historical control range of the testing facility,

and all fetuses with cleft palate had lower body weights. Cleft palates are likely to occur with dams with stress-induced changes and fetal growth retardation (*J Exp Zool.* 1957; 134: 449-67). Taking into account that reduced maternal body weight gain, suggestive of maternal stress, was observed in the 300 mg/kg/day group, cleft palate may have occurred as a secondary consequence of either maternal toxicity itself or reduced fetal body weight resulting from maternal toxicity. Because no effects on embryo-fetal development were observed in the 100 mg/kg/day group, the NOAEL for embryo-fetal development in mice was determined to be 100 mg/kg/day.

- In addition to the above studies, given that fetal malformations reported in some of embryo-fetal developmental toxicity studies of other compounds that enhance GABA_A receptor function like zuranolone were not consistent, the embryo-fetal effects of zuranolone observed in rats and mice were unlikely related to enhancement of GABA_A receptor function. Thus, the embryo-fetal effects of zuranolone observed in rats were likely secondary to maternal toxicity or were likely off-target toxicity, and the embryo-fetal effects of zuranolone observed in mice were likely secondary to maternal toxicity.

Then, the use of zuranolone in pregnant women or women who may be pregnant is not recommended because fetal malformations for which a relationship to zuranolone could not be ruled out were observed in the embryo-fetal developmental toxicity study in rats. However, for the following reasons etc., the package insert should advise, instead of a contraindication, that it is desirable not to administer zuranolone.

- Since zuranolone exposure ($AUC_{0-last} = 9380 \text{ ng}\cdot\text{h/mL}$) at the NOAEL of 7.5 mg/kg/day for embryo-fetal development in rats was approximately 10-fold the human exposure ($AUC_{0-24h} = 943.7 \text{ ng}\cdot\text{h/mL}$) following oral administration of the clinical dose (30 mg/day) of zuranolone, the findings observed in the embryo-fetal developmental toxicity study raise no serious concerns about human use.
- Cleft palates observed in the embryo-fetal developmental toxicity study in mice were not considered a direct effect of zuranolone on embryo-fetal development.
- In addition to the above discussion on the results of the embryo-fetal developmental toxicity studies, zuranolone that can provide a rapid response as compared with the currently approved antidepressants such as selective serotonin reuptake inhibitors (SSRIs) may contribute to reductions in the suicides of pregnant women and the risk to infant development. Given that suicide associated with depression has become a problem as the cause of death of pregnant women and nursing mothers (Japan Society of Obstetrics and Gynecology, Japan Suicide Countermeasures Promotion Center¹⁷⁾), the use of zuranolone in pregnant women has a certain significance.

PMDA's view on the embryo-fetal effects of zuranolone observed in the embryo-fetal developmental toxicity studies in rats and mice (Table 17):

Absent rib, fused rib, and fused thoracic arch observed at $\geq 7.5 \text{ mg/kg/day}$ in the embryo-fetal developmental toxicity study in rats occur only very rarely as spontaneous findings (*Congenit Anom (Kyoto)*. 2019; 59: 125-31), and the incidences of these findings tended to increase dose-dependently. In addition, severe maternal toxicity such as severe sedation was not observed at 7.5 mg/kg/day. Taking account of these points etc., the

17) <https://jscp.or.jp/assets/img/maternal-suicide.pdf> (as of August 29, 2025)

possibility that zuranolone had a direct effect on embryos/fetuses cannot be ruled out. Then, given that the incidences of multiple malformations observed in the 22.5 mg/kg/day group were higher than the historical control range of the testing facility and published literature values (*Congenit Anom (Kyoto)*. 2019; 59: 125-31), these study results indicate that zuranolone has a teratogenic risk. Based on the above, the NOAEL for embryo-fetal developmental toxicity in rats should be 2.5 mg/kg/day.

As explained by the applicant, cleft palates observed in the embryo-fetal developmental toxicity study in mice were considered secondary to maternal toxicity or lower fetal body weights. The NOAEL for embryo-fetal development in mice of 100 mg/kg/day is appropriate.

Based on the above, PMDA's view on the use of zuranolone in pregnant women or women who may be pregnant:

Given that zuranolone exposures ($AUC_{0-last} = 3710 \text{ ng}\cdot\text{h/mL}$ in rats, $AUC_{0-last} = 5500 \text{ ng}\cdot\text{h/mL}$ in mice) at 2.5 mg/kg/day that did not result in embryo-fetal developmental toxicity, which is considered the NOAEL for embryo-fetal development in rats, and at the NOAEL for embryo-fetal development in mice of 100 mg/kg/day were 3.9-fold and 5.8-fold the AUC_{0-24h} ($943.7 \text{ ng}\cdot\text{h/mL}$) in humans following oral administration of the clinical dose (30 mg/day) of zuranolone, the possibility that zuranolone in clinical use induces developmental toxicity including teratogenicity cannot be ruled out. Thus, the use of zuranolone in pregnant women or women who may be pregnant is not recommended, and taking account of the risk of zuranolone to the embryo/fetus, the efficacy of zuranolone in the treatment of depressive symptoms, and other treatment options for depressive symptoms in Japan, it is difficult to conclude that the benefits of administering zuranolone to patients with depression outweigh the risk of zuranolone to the embryo/fetus. Thus, the package insert should state that zuranolone is contraindicated in pregnant women or women who may be pregnant and should include a relevant precautionary statement.

5.R.2 Contraception in women of reproductive potential and male patients of reproductive potential

Given that embryo-fetal developmental toxicity associated with zuranolone was observed [see Section 5.R.1], PMDA asked the applicant to explain contraception in women of reproductive potential and male patients of reproductive potential.

The applicant's explanation:

Given that the half-life of zuranolone is approximately 15 to 20 hours [see Section 6.2.2.1], women of reproductive potential should use effective contraception during treatment with zuranolone and for 1 week after the final dose to avoid unintended embryo/fetal exposure to zuranolone.

As to contraception in male patients of reproductive potential, if zuranolone is excreted into semen following administration of zuranolone in a male patient, and then zuranolone is 100% absorbed into the circulation through the vaginal mucosa of a female partner, the plasma exposure of zuranolone (AUC) in the female partner

is estimated to be 0.169 ng·h/mL,¹⁸⁾ which is far from plasma exposures ($AUC_{0-last} = 9380$ ng·h/mL in rats, $AUC_{0-last} = 5500$ ng·h/mL in mice) at the NOAELs for embryo-fetal development in rats and mice (7.5 mg/kg/day in rats, 100 mg/kg/day in mice). Thus, zuranolone is unlikely to affect the embryo/fetus via semen, and contraception in male patients is unnecessary.

PMDA's conclusion:

The applicant's explanation (The package insert should advise that female patients of reproductive potential should use effective contraception during treatment with zuranolone and for 1 week after the final dose, and contraception in male patients is unnecessary.) is appropriate.

5.R.3 Use in lactating women

Zuranolone has been shown to be excreted in human milk [see Section 6.2.2.3]. Decreased body weights of pre-weaning F₁ pups were observed in a rat study for effects on pre- and postnatal development, including maternal function (CTD 4.2.3.5-07) (Table 17). Activation of GABA_A receptors has been reported to trigger neurodegeneration in infant rats (*Proc Natl Acad Sci USA*. 2002; 99: 15089-94). Based on these findings, a neurodegeneration study in juvenile animals (CTD 4.2.3.5-10) was conducted. PMDA asked the applicant to explain whether breastfeeding should be continued or discontinued during treatment with zuranolone.

The applicant's explanation:

For the following reasons etc., zuranolone is unlikely to affect nursing infants via milk. Thus, the package insert should advise that a decision to continue or discontinue breastfeeding should be made, taking account of the therapeutic benefits and the benefits of breastfeeding nutrition.

- In the rat study for effects on pre- and postnatal development, including maternal function, decreased body weights of pre-weaning F₁ pups were observed only at the doses associated with decreased food consumption in dams (≥ 4 mg/kg/day) (Table 17). Given that body weights of F₁ pups were reduced due to maternal undernutrition during pregnancy in rats (*Reprod Toxicol*. 2009; 28: 489-94), decreased body weights of pre-weaning F₁ pups were considered related to reduced food consumption in dams. Zuranolone exposure ($AUC_{0-last} = 2420$ ng·h/mL) at 1 mg/kg/day that did not result in F₁ pup effects was 2.56-fold the human exposure at the clinical dose ($AUC_{0-24h} = 943.7$ ng·h/mL).
- The infant dose of zuranolone via human milk was 0.00125 mg/kg/day [see Section 6.2.2.3]. When compared with the human equivalent dose calculated from the NOEL (2.5 mg/kg/day) in the neurodegeneration study in juvenile rats (Table 18), there is an approximately 250-fold safety margin.

PMDA's view:

Although decreased body weights of pups were observed in the rat study for effects on pre- and postnatal development, including maternal function, given the above explanation by the applicant, decreased body

18) Based on a conservative estimate of 6 for the semen to plasma zuranolone concentration ratio made from a tissue distribution study in rats (CTD 4.2.2.2-16), the C_{max} (67.2 ng/mL) in a male patient, and a semen volume per ejaculation (6 mL), the dose to which the vagina of a female partner is exposed was calculated ($67.2 \text{ ng/mL} \times 6 \text{ mL} \times 6 = 0.00242 \text{ mg}$). Using this dose, assuming that zuranolone is 100% absorbed into the circulation through the vaginal mucosa and given zuranolone exposure ($AUC_{0-24h} = 943.7$ ng·h/mL) at the clinical dose of 30 mg and the oral absorption rate of zuranolone of at least 45% based on the results from a mass balance study (CTD 5.3.3.1-02), plasma exposure (AUC) in the female partner was calculated ($943.7 \text{ ng·h/mL} \times (0.00242/30) \times (100/45) = 0.169 \text{ ng·h/mL}$).

weights of pups were considered related to decreased food consumption in dams and were unlikely to be a direct effect of zuranolone on suckling rats via milk. In addition, taking also into account that decreased body weights of pups observed in this study were 93% of the control group, there should be little concern about the effect of zuranolone on nursing infants via milk. Thus, the applicant's explanation about the use of zuranolone in lactating women (The package insert should advise that a decision to continue or discontinue breastfeeding should be made, taking account of the therapeutic benefits and the benefits of breastfeeding nutrition.) is appropriate.

6. Summary of Biopharmaceutic Studies and Associated Analytical Methods, Clinical Pharmacology, and Outline of the Review Conducted by PMDA

6.1 Summary of biopharmaceutic studies and associated analytical methods

Zuranolone concentrations in biomaterials were determined using LC-MS/MS (LLOQ, 1.00 ng/mL).

The to-be-marketed formulation was used in the pivotal clinical studies of zuranolone, i.e., Japanese phase III studies (Studies A3734 and A3736).

6.1.1 Food effect (CTD 5.3.3.1-01, Study A3711 [20 to 20])

In Part B of Study A3711 [for the parts of Study A3711, see Section 7.1.1], Japanese healthy adult subjects (12 subjects included in pharmacokinetic assessment) received a single oral dose of the zuranolone white capsule¹⁹⁾ 30 mg under fasted conditions or after ingestion of a high-fat meal (≥ 900 kcal, $\geq 35\%$ fat) in a crossover manner to assess the effect of food on the pharmacokinetics of zuranolone. Table 20 shows the pharmacokinetic parameters of zuranolone under fasted conditions or after ingestion of a high-fat meal.

Table 20. Plasma pharmacokinetic parameters of zuranolone after administration of zuranolone under fasted conditions or after ingestion of high-fat meal

Food	N	C _{max} (ng/mL)	t _{max} (h) ^{a)}	AUC _{0-last} (ng·h/mL)	Geometric mean ratio [90% CI] (High-fat meal/fasted)	
					C _{max}	AUC _{0-last}
Fasted	12	18.3 (50.2)	4.0 (2.0, 10.0)	352.9 (38.1)	4.09	2.37
High-fat meal	12	74.8 (17.1)	5.0 (3.0, 8.0)	836.2 (15.1)	[3.39, 4.94]	[2.02, 2.79]

Geometric mean (CV [%])

a) Median (Min., Max.)

6.2 Clinical pharmacology

6.2.1 Studies using human biomaterials

(1) Plasma protein binding (Reference data CTD 5.3.2.1-01, 5.3.2.1-03)

When zuranolone (10 μ mol/L) was added to human plasma, the plasma protein binding of zuranolone as determined by an equilibrium dialysis method was 98.8%.

When zuranolone (30-500 ng/mL) was added to α 1-acid glycoprotein (1.0 mg/mL) or human serum albumin (45 mg/mL), the binding of zuranolone to α 1-acid glycoprotein and human serum albumin was 89.7% to 91.6% and 99.3% to 99.4%, respectively, both of which were independent of the zuranolone concentration.

¹⁹⁾ This formulation differs in capsule shell color only from the to-be-marketed formulation.

(2) Identification of metabolites in humans (Reference data CTD 5.3.2.2-08)

When zuranolone 100 µmol/L was added to human cryopreserved hepatocytes and incubated at 37°C for 240 minutes, M1 to M4 (mono-hydroxylated metabolites) were formed. Although relative abundance of metabolites was not determined, HPLC chromatograms suggest that the most abundant metabolite was M4 and that M1 was present in trace amounts.

(3) Identification of enzymes involved in metabolism (Reference data CTD 5.3.2.2-01)

In the presence or absence of inhibitors of cytochrome P450 isoforms,²⁰⁾ zuranolone (2 µmol/L) was incubated with human liver microsomes. CYP3A inhibitor produced 50% reduction in the formation of M1 and 96% to 97% reductions in the formation of M2 to M4. CYP2B6, CYP2C8, and CYP2C9 inhibitors produced 22% to 25%, 35% to 48%, and 22% to 33% reductions in the formation of M1 to M4, respectively. CYP1A2, CYP2C19, or CYP2D6 inhibitor caused no evident inhibition of metabolism of zuranolone. The applicant explained that the above results indicate that zuranolone is mainly metabolized by CYP3A.

(4) Enzyme inhibition or induction

Using the substrates for CYP1A2, CYP2C9, CYP2C19, CYP2D6, and CYP3A,²¹⁾ the potential for zuranolone (0.003-3 µmol/L for CYP1A2, CYP2C9, CYP2C19, and CYP2D6, 0.03-30 µmol/L for CYP3A) to inhibit the different cytochrome P450 isoforms in human liver microsomes was investigated. Zuranolone was a reversible inhibitor of CYP3A (IC_{50} = 16-29 µmol/L). No evident inhibition was noted for other CYP isoforms within the concentration range tested (IC_{50} >3 µmol/L). Zuranolone did not cause time- or metabolism-dependent inhibition of any of the CYP isoforms tested (CTD 5.3.2.2-03, Study SSN-02183).

Using the substrates for CYP2B6 and CYP2C8,²²⁾ the potential for zuranolone (0.02-20 µmol/L) to inhibit the different CYP isoforms in human liver microsomes was investigated. Zuranolone was a reversible inhibitor of CYP2C8 (IC_{50} = 14 µmol/L). No evident inhibition was noted for CYP2B6 within the concentration range tested (IC_{50} >20 µmol/L). Zuranolone did not cause time- or metabolism-dependent inhibition of CYP2B6 or CYP2C8 (CTD 5.3.2.2-02, Study SSN-01539).

The applicant's explanation:

Given the above results, the C_{max} (67.2 ng/mL)¹⁰⁾ following oral administration of the clinical dose (30 mg/day) of zuranolone, and the plasma protein binding of zuranolone, zuranolone is unlikely to cause drug interactions via cytochrome P450 inhibition.

Using the substrates for CYP1A2, CYP2B6, CYP2C8, CYP2C9, CYP2C19, CYP2D6, and CYP3A,²³⁾ the

20) furaflavone 10 µmol/L for CYP1A2, thiotepa 50 µmol/L for CYP2B6, montelukast 3 µmol/L for CYP2C8, sulfaphenazole 10 µmol/L for CYP2C9, benzylnirvanol 1 µmol/L for CYP2C19, quinidine 1 µmol/L for CYP2D6, diethyldithiocarbamate 50 µmol/L for CYP2E1, ketoconazole 1 µmol/L for CYP3A

21) phenacetin for CYP1A2, diclofenac for CYP2C9, S-mephenytoin for CYP2C19, dextromethorphan for CYP2D6, midazolam and testosterone for CYP3A

22) efavirenz for CYP2B6, amodiaquine for CYP2C8

23) phenacetin for CYP1A2, efavirenz for CYP2B6, amodiaquine for CYP2C8, diclofenac for CYP2C9, S-mephenytoin for CYP2C19, dextromethorphan for CYP2D6, midazolam and testosterone for CYP3A

potential for zuranolone metabolites,⁷⁾ M117 (0.01-10 µmol/L), M125 (0.01-10 µmol/L), M135 (0.015-15 µmol/L), and M136 (0.015-15 µmol/L), to inhibit the different cytochrome P450 isoforms in human liver microsomes was investigated. M117 was a reversible inhibitor of CYP2C8 (IC_{50} = 6.3 µmol/L), and no evident inhibition was noted for other cytochrome P450 isoforms within the concentration range tested (IC_{50} >10 µmol/L). It was not a metabolism-dependent inhibitor of any of the cytochrome P450 isoforms tested. M125 exhibited no evident reversible inhibition of any of the cytochrome P450 isoforms tested (IC_{50} >10 µmol/L) and was not a metabolism-dependent inhibitor. M135 was a reversible inhibitor of CYP2B6, CYP2C8, CYP2C19, and CYP2D6 (IC_{50} values were 14, 3.9, 13, and 0.43 µmol/L, respectively), and no evident inhibition was noted for CYP1A2, CYP2C9, and CYP3A within the concentration range tested (IC_{50} >15 µmol/L). Although M135 showed a trend towards metabolism-dependent inhibition of CYP3A, the IC_{50} value was >15 µmol/L. M136 was a reversible inhibitor of CYP2C8 (IC_{50} = 8.9 µmol/L), and no evident inhibition was noted for other cytochrome P450 isoforms within the concentration range tested (IC_{50} >15 µmol/L). It was a metabolism-dependent inhibitor of CYP3A (IC_{50} >13-15 µmol/L). There was no evidence of time-dependent inhibition in any of the cytochrome P450 isoforms evaluated by any of the metabolites (CTD 5.3.2.2-04, Study SSN-03597 and CTD 5.3.2.2-05, Study SSN-03598).

The applicant's explanation:

Given the above results, the C_{max} values of M117, M125, M135, and M136 determined in a mass balance study, and the plasma protein binding of these metabolites,²⁴⁾ these metabolites are unlikely to cause drug interactions via cytochrome P450 inhibition.

Zuranolone (0.1-30 µmol/L) was added to human hepatocytes, and the potential for zuranolone to induce CYP1A2, CYP2B6, and CYP3A4 was investigated based on mRNA expression and catalytic activity. CYP2B6 mRNA and activity increased in an approximately concentration-dependent manner, with maximal 3.8-fold and 4.0-fold induction (at 10 µmol/L and 30 µmol/L of zuranolone, respectively) (17% and 31% of positive control [phenobarbital 1000 µmol/L] response, respectively). CYP3A4 mRNA increased in an approximately concentration-dependent manner, with maximal 6.1-fold induction (at 30 µmol/L of zuranolone) (31% of positive control [rifampicin 10 µmol/L] response), whereas no induction of CYP3A4 activity was observed within the concentration range tested. Zuranolone was not an inducer of CYP1A2 mRNA or activity within the concentration range tested (Reference data CTD 5.3.2.2-06, Study SSN-01208).

Based on the above results, the CYP2B6 and CYP3A4 induction potential of zuranolone was evaluated in clinical drug interaction studies (Table 26).

(5) Transporter substrate potential (CTD 5.3.2.2-07, Study SSN-02185)

The cell permeability of zuranolone (1-100 µmol/L) was assessed in Madin-Darby canine kidney (MDCK) II cells expressing breast cancer resistance protein (BCRP) or P-glycoprotein (P-gp). As the Net ER²⁵⁾ of

24) The plasma protein binding of M117 (0.5-5 µmol/L), M125 (0.5-5 µmol/L), M135 (2-10 µmol/L), and M136 (2-10 µmol/L) was 99.8%-100%, 98.3%-99.3%, 95.0%-99.3%, and 95.2%-99.0%, respectively (CTD 5.3.2.1-02).

25) Efflux ratio in P-gp- or BCRP-expressing cells – efflux ratio in parental cells

zuranolone was <1 , zuranolone was not considered a substrate of P-gp or BCRP.

Using human embryonic kidney 293 (HEK293) cell line expressing organic anion transporting polypeptide 1B1 (OATP1B1) or OATP1B3, each transporter-mediated cellular uptake of zuranolone (0.3 and 3 $\mu\text{mol/L}$) was investigated in the presence or absence of the respective transporter inhibitor (rifampicin 50 $\mu\text{mol/L}$). There were no clear differences in the uptake of zuranolone in the presence or absence of the inhibitor. Thus, zuranolone was not considered a substrate of OATP1B1 or OATP1B3.

(6) Transporter inhibition potential (CTD 5.3.2.2-07, Study SSN-02185)

Using HEK-293 cells expressing OATP1B1, OATP1B3, organic anion transporter 1 (OAT1), OAT3, organic cation transporter 1 (OCT1), or OCT2, MDCKII cells expressing multidrug and toxin extrusion protein 1 (MATE1) or MATE2-K, or HEK-293 membrane vesicles expressing human bile salt export pump (BSEP), BCRP, or P-gp, the potential for zuranolone (0.3 or 3 $\mu\text{mol/L}$ for OATP1B1, OAT3, and BSEP, 0.25 or 2.5 $\mu\text{mol/L}$ for OATP1B3, OAT1, OCT1, OCT2, MATE1, and MATE2-K, 0.63 or 6.26 $\mu\text{mol/L}$ for BCRP and P-gp) to inhibit the transport of the substrates of these transporters²⁶⁾ was investigated. Zuranolone caused no evident inhibition of any of the transporters within the concentration range tested (IC_{50} values were >3 $\mu\text{mol/L}$ for OATP1B1, OAT3, and BSEP, >2.5 $\mu\text{mol/L}$ for OATP1B3, OAT1, OCT1, OCT2, MATE1, and MATE2-K, and >6.26 $\mu\text{mol/L}$ for BCRP and P-gp).

The applicant's explanation:

Given the above results, the C_{max} (67.2 ng/mL)¹⁰⁾ following oral administration of the clinical dose (30 mg/day) of zuranolone, and the plasma protein binding of zuranolone, zuranolone is unlikely to cause drug interactions via transporter inhibition.

6.2.2 Healthy adult subject studies

6.2.2.1 Japanese phase I study (CTD 5.3.3.1-01, Study A3711 [20 to 20])

Japanese and non-Japanese (Caucasian) healthy adult subjects (54 subjects included in pharmacokinetic assessment [27 non-elderly Japanese subjects, 18 non-elderly non-Japanese subjects, and 9 Japanese elderly subjects]) received a single oral dose of zuranolone 10, 20, or 30 mg or zuranolone 10, 20, or 30 mg once daily for 7 days under fed conditions [for the details of dosing regimen, see Section 7.1.1].

26) estradiol-17 β -D-glucuronide for OATP1B1, cholecystokinin-8 for OATP1B3, tenofovir for OAT1, estrone-3-sulfate for OAT3 and BCRP, metformin for OCT1, OCT2, MATE1, and MATE2-K, taurocholic acid for BSEP, N-methyl-quinidine for P-gp

Table 21 shows the plasma pharmacokinetic parameters of zuranolone after a single dose, and Table 22 shows the plasma pharmacokinetic parameters of zuranolone after 7 days of dosing. Although the C_{max} and AUC at 20 mg of zuranolone tended to be higher in Japanese subjects than in non-Japanese subjects, there were no clear differences in the C_{max} and AUC at 30 mg between Japanese and non-Japanese subjects. There were no clear differences in the pharmacokinetics of zuranolone between non-elderly and elderly Japanese subjects.

Table 21. Plasma pharmacokinetic parameters of zuranolone after a single oral dose of zuranolone in healthy adult subjects

	Dose (mg)	Number of evaluable subjects	C_{max} (ng/mL)	t_{max} (h) ^{a)}	AUC _{0-last} (ng·h/mL)	$t_{1/2}$ (h)
Non-elderly Japanese subjects	10	9	27.6 (32.5)	5.0 (4.0, 6.0)	255.0 (43.8)	13.5 (36.9)
	20	9	67.8 (22.4)	5.0 (4.0, 6.0)	717.9 (23.5)	14.1 (29.5)
	30	9	80.3 (18.9)	5.0 (4.0, 10.0)	911.2 (17.1)	13.9 (26.6)
Non-elderly non-Japanese subjects	20	9	50.7 (32.4)	5.0 (3.0, 8.0)	497.5 (34.7)	18.7 (51.6)
	30	9	72.7 (18.6)	5.0 (3.0, 5.0)	898.8 (32.9)	20.2 (35.6)
Japanese elderly subjects	30	9	76.4 (16.8)	5.0 (4.0, 5.0)	799.0 (27.6)	17.9 (24.5)

Geometric mean (CV [%])

a) Median (Min., Max.)

Table 22. Plasma pharmacokinetic parameters of zuranolone after multiple oral doses of zuranolone in healthy adult subjects

	Dose (mg)	Number of evaluable subjects	C_{max} (ng/mL)	t_{max} (h) ^{a)}	AUC _{0-τ} (ng·h/mL)	$t_{1/2}$ (h)
Non-elderly Japanese subjects	10	9	18.9 (32.2)	4.0 (4.0, 12.0)	227.1 (29.5)	16.3 (27.8)
	20	9	39.3 (30.8)	12.0 (3.0, 12.0)	556.6 (24.4)	16.4 (23.9)
	30	9	48.6 (22.4)	12.0 (4.0, 12.0)	723.0 (20.5)	15.4 (20.8)
Non-elderly non-Japanese subjects	20	9	23.8 (35.8)	12.0 (4.0, 12.0)	365.4 (42.6)	21.3 (30.5)
	30	9	49.9 (25.4)	12.0 (4.0, 12.0)	837.2 (24.6)	23.9 (37.3)
Japanese elderly subjects	30	9	54.7 (30.0)	4.0 (4.0, 12.0)	804.3 (28.2)	20.2 (19.8)

Geometric mean (CV [%])

a) Median (Min., Max.)

6.2.2.2 Mass balance study in non-Japanese healthy adult subjects (CTD 5.3.3.1-02, 5.3.2.2-12, Study 217-CLP-105 [■ to ■ 20■])

The mass balance of a single oral dose of ¹⁴C-zuranolone 30 mg under fasted conditions was determined in non-Japanese healthy adult subjects (8 subjects included in pharmacokinetic assessment). In the plasma collected up to 336 hours post-dose, unchanged zuranolone was detected, and the AUC_{0-last} of unchanged zuranolone accounted for 4.9% of the total radioactivity in plasma. The plasma AUC_{0-last} values of metabolites M63 (structurally unidentified) and M125 were higher than that of unchanged zuranolone, and the AUC_{0-last} values of M63 and M125 accounted for 6.4% and 7.5% of the total radioactivity in plasma, respectively. The blood to plasma partitioning ratio up to 24 hours post-dose was 0.54 to 0.58.

Up to 144 hours post-dose, 41.2% of the administered radioactivity was recovered in urine, and unchanged zuranolone was not detected in urine. Up to 216 hours post-dose, 37.0% of the administered radioactivity was recovered in feces, and 1.6% of the administered radioactivity was recovered as unchanged zuranolone in feces.

6.2.2.3 Phase I study in non-Japanese lactating women (CTD 5.3.3.1-03, Study 217-CLP-114 [■ to ■ 20■])

Non-Japanese healthy adult lactating women (14 subjects included in pharmacokinetic assessment) received oral zuranolone 30 mg once daily after the evening meal for 5 days. Based on the zuranolone concentrations

in milk on Day 5, the mean daily infant dose of zuranolone²⁷⁾ via milk at the steady-state plasma concentration of zuranolone was calculated to be 0.00125 mg/kg/day.

6.2.3 Intrinsic factor pharmacokinetic studies

6.2.3.1 Pharmacokinetic study in subjects with renal impairment (CTD 5.3.3.3-01, Study 217-CLP-107 [20 to 20])

Table 23 shows the plasma pharmacokinetic parameters of zuranolone in non-Japanese healthy adult subjects (6 subjects with normal renal function with an estimated glomerular filtration rate (eGFR) ≥ 90 mL/min/1.73 m²) and subjects with renal impairment (6 subjects with mild impairment [eGFR of 60-89 mL/min/1.73 m²], 5 subjects with moderate impairment [eGFR of 30-59 mL/min/1.73 m²], 7 subjects with severe impairment [eGFR < 30 mL/min/1.73 m²]) after a single oral dose of zuranolone 30 mg under fed conditions. At 5 hours post-dose, the fraction of zuranolone unbound to plasma proteins (geometric mean) in subjects with normal renal function or mild, moderate, or severe renal impairment was 0.32%, 0.28%, 0.31%, and 0.23%, respectively, showing no clear differences according to the degree of renal impairment.

Table 23. Plasma pharmacokinetic parameters of zuranolone after a single dose of zuranolone 30 mg in subjects with normal renal function or renal impairment

Renal function	Dose (mg)	Number of evaluable subjects	C _{max} (ng/mL)	t _{max} (h) ^{a)}	t _{1/2} (h)	AUC _{0-∞} (ng·h/mL)	Geometric least-squares mean ratio [90% CI]	
							C _{max}	AUC _{0-∞}
Normal	30	6	47.9 (39.8)	5.0 (2.0, 6.0)	22.0 (26.5)	730.1 (51.6)		
Mild impairment	30	6	59.6 (19.5)	5.5 (4.0, 8.0)	26.9 (14.5)	948.2 (19.1)	1.24 [0.91, 1.71]	1.30 [0.88, 1.91]
Moderate impairment	30	5	44.0 (69.6)	5.0 (3.0, 6.0)	28.7 (20.4)	1091 (21.9)	0.92 [0.52, 1.61]	1.50 [0.97, 2.30]
Severe impairment	30	7	49.1 (28.8)	6.0 (6.0, 12.0)	29.6 (39.3)	1021 (35.3)	1.03 [0.74, 1.43]	1.40 [0.92, 2.12]

Geometric mean (CV [%])

a) Median (Min., Max.)

6.2.3.2 Pharmacokinetic study in subjects with hepatic impairment (CTD 5.3.3.3-02, Study 217-CLP-108 [20 to 20])

Table 24 shows the plasma pharmacokinetic parameters of zuranolone in non-Japanese healthy adult subjects (6 subjects with normal hepatic function) and subjects with hepatic impairment (mild impairment [Child-Pugh Class A], moderate impairment (Child-Pugh Class B), and severe impairment [Child-Pugh Class C] [6 subjects each]) after a single oral dose of zuranolone 30 mg (zuranolone 20 mg for subjects with severe hepatic impairment) under fed conditions. At 5 hours post-dose, the fraction of zuranolone unbound to plasma proteins (geometric mean) in subjects with normal hepatic function or mild, moderate, or severe hepatic impairment was 0.24%, 0.16%, 0.29%, and 0.26%, respectively, showing no clear differences according to the degree of hepatic impairment.

27) (Total amount of drug calculated from milk volume in each milk collection and drug concentration in each milk collection over a 24-hour period)/(baseline infant weight)

Table 24. Plasma pharmacokinetic parameters of zuranolone after a single dose of zuranolone 30 or 20 mg in subjects with normal hepatic function or hepatic impairment

Hepatic function	Dose (mg)	Number of evaluable subjects	C _{max} (ng/mL)	t _{max} (h) ^{a)}	t _{1/2} (h)	AUC _{0-∞} (ng·h/mL)	Geometric least-squares mean ratio for dose-normalized parameter [90% CI]	
							C _{max}	AUC _{0-∞}
Normal	30	6	60.6 (21.5)	5.0 (3.0, 8.0)	24.58 (26.8)	1044 (29.3)		
Mild impairment	30	6	59.2 (31.4)	5.0 (5.0, 12.0)	22.40 (38.9)	887.5 (45.4)	0.99 [0.77, 1.26]	0.86 [0.58, 1.28]
Moderate impairment	30	6	54.7 (30.4)	4.5 (3.0, 6.0)	30.41 (36.9)	1020 (55.6)	0.97 [0.78, 1.20]	1.08 [0.74, 1.58]
Severe impairment	20	6	28.7 (19.1)	5.0 (3.0, 8.0)	43.96 (44.7)	1074 (41.5)	0.76 [0.58, 0.98]	1.56 [0.98, 2.48]

Geometric mean (CV (%))

a) Median (Min., Max.)

6.2.4 Drug interaction studies (CTD 5.3.3.4-01, Study 217-CLP-102 [■ to ■ 20■]; CTD 5.3.3.4-02, Study 217-CLP-106 [■ to ■ 20■])

The drug-drug interaction potential of zuranolone²⁸⁾ was evaluated. Table 25 and Table 26 show the geometric mean ratios of the plasma pharmacokinetic parameters of zuranolone or the coadministered drug when administered concomitantly relative to administration alone.

Table 25. Effect of coadministered drug on plasma pharmacokinetics of zuranolone

Coadministered drug (Dosing regimen)	Dosing regimen of zuranolone	Number of evaluable subjects	Geometric mean ratio ^{a)} [90% CI]		Study ID CTD
			C _{max}	AUC _{0-∞}	
Rifampicin 600 mg QD ^{b)}	30 mg single dose	16	0.31 [0.27, 0.37]	0.15 [0.13, 0.17]	217-CLP-106 5.3.3.4-02
Itraconazole 200 mg QD ^{c)}	20 mg single dose	16	1.25 [1.13, 1.39]	1.62 [1.54, 1.70]	

a) Coadministration/administration alone

b) Two single oral doses of zuranolone 30 mg under fed conditions on Days 1 and 11 with multiple oral doses of rifampicin 600 mg QD on Days 4 to 10

c) Two single oral doses of zuranolone 20 mg under fed conditions on Days 1 and 9 with multiple oral doses of itraconazole 200 mg QD on Days 5 to 12

Table 26. Effect of zuranolone on plasma pharmacokinetics of coadministered drug

Dosing regimen of zuranolone	Coadministered drug (Dosing regimen)	Analyte	Number of evaluable subjects	Geometric mean ratio ^{a)} [90% CI]		Study ID CTD
				C _{max}	AUC _{0-∞}	
30 mg QD ^{b)}	Bupropion (100 mg QD)	Bupropion	12	0.92 [0.77, 1.11]	1.04 [0.96, 1.14]	217-CLP-102 5.3.3.4-01
		Hydroxyl-bupropion	12	0.85 [0.78, 0.92]	0.87 [0.81, 0.94] ^{c)}	
	Simvastatin (20 mg QD)	Simvastatin	12	1.10 [0.85, 1.43]	1.13 [1.04, 1.23]	
		Simvastatin acid	12	1.25 [0.91, 1.73]	1.11 [0.75, 1.64] ^{d)}	

a) Coadministration/administration alone

b) Bupropion 100 mg on Days 1 and 11 and simvastatin 20 mg on Days 2 and 10 with multiple oral doses of zuranolone 30 mg QD under fasted conditions on Days 3 to 9

c) AUC_{0-last}

d) N = 10

6.2.5 Pharmacodynamic study

6.2.5.1 QT/QTc evaluation study (CTD 5.3.4.1-01, Study 217-CLP-112 [■ to ■ 20■])

A placebo- and active-controlled, randomized, double-blind, parallel-group study using moxifloxacin as a positive control was conducted in non-Japanese healthy adult subjects (64 subjects included in pharmacodynamic assessment) to evaluate the effect of zuranolone on QT/QTc interval. Table 27 shows the dosing schedules of placebo, zuranolone, or moxifloxacin, all of which were administered orally once daily

28) In Study 217-CLP-102, zuranolone oral solution, i.e., zuranolone formulated in 40% (w/v) HPβCD aqueous solution, was used.

under fed conditions.

Table 27. Dosing schedules

Treatment group	N	Day 1	Day 2	Days 3-7	Day 8	Day 9
Group 1	32	Placebo	Zuranolone 50 mg	Zuranolone 50 mg	Zuranolone 100 mg	Placebo
Group 2A	16	Placebo	Moxifloxacin 400 mg	Placebo	Placebo	Placebo
Group 2B	16	Placebo	Placebo	Placebo	Placebo	Moxifloxacin 400 mg

The relationship between the plasma zuranolone concentration and the change from baseline in the QT interval corrected for heart rate using Fridericia's method (ΔQTcF) was analyzed using a pre-specified linear mixed-effects model. At zuranolone exposure after oral administration of zuranolone 100 mg (the geometric mean C_{max} = 132.8 ng/mL [higher than 97.4 ng/mL,²⁹⁾ i.e., the expected highest exposure after administration of the clinical dose (in the worst case scenario)], the upper bound of the 90% confidence interval (CI) for the placebo-corrected change from baseline in the QT interval ($\Delta\Delta\text{QTcF}$) was 5.1 msec and was estimated to be below 10 msec. Thus, the risk of QT interval prolongation associated with zuranolone should be low. The relationship between the plasma moxifloxacin concentration and ΔQTcF was analyzed using a linear mixed-effects model. The lower bound of the 90% CI for $\Delta\Delta\text{QTcF}$ after administration of moxifloxacin 400 mg was estimated to be 7.0 msec.

6.2.5.2 Human abuse potential study (Reference data CTD 5.3.4.1-02, Study 217-CLP-110 [■ to ■ 20■])

Non-Japanese subjects who had used recreational CNS depressants (60 subjects included in pharmacodynamic assessment) received a single oral dose of placebo, zuranolone 30, 60, or 90 mg, or alprazolam 1.5 or 3 mg (positive control) under fed conditions in a crossover fashion. The maximum effect (E_{max}) of the Drug Liking VAS³⁰⁾ (mean $\pm$ standard deviation [SD]) was 58.1 ± 12.8 for placebo, 68.5 ± 16.7 , 76.3 ± 14.4 , and 83.3 ± 12.9 for zuranolone 30, 60, and 90 mg, respectively, and 79.8 ± 13.5 and 83.7 ± 12.2 for alprazolam 1.5 and 3 mg, respectively. The E_{max} of the Drug Liking VAS was higher for zuranolone 30 and 60 mg than for placebo, but lower as compared with alprazolam 1.5 and 3 mg. There were no statistically significant differences between zuranolone 90 mg and alprazolam 1.5 or 3 mg.

6.2.5.3 Driving study (Reference data CTD 5.3.4.1-03, Study 217-CLP-113 [■ to ■ 20■])

Placebo, zuranolone 30 mg or 30/60 mg,³¹⁾ or zopiclone 7.5 mg was administered orally once daily after the evening meal for 5 days in a crossover fashion to non-Japanese healthy adult subjects (60 subjects included in pharmacodynamic assessment), and the effect of zuranolone on driving ability was assessed using a driving simulator. The placebo-corrected mean Standard Deviation of Lateral position (SDLP)³²⁾ in the morning on Day 2 following a single dose of zuranolone 30 mg or zopiclone 7.5 mg [95% CI] was 3.14 [1.56, 4.71]³³⁾ or

29) The predicted C_{max} after repeated oral administration of zuranolone 30 mg/day with a strong CYP3A4 inhibitor. The PPK model-predicted C_{max} after repeated oral administration of zuranolone 30 mg/day was 67.2 ng/mL, and the results of Study 217-CLP-106 (CTD 5.3.3.4-02) indicated that repeated oral administration of zuranolone with a strong CYP3A inhibitor increases the C_{max} by 1.45-fold.

30) Drug liking was measured using VAS (0 [strongly disliking] to 100 [strongly liking]).

31) Zuranolone 30 mg was administered on Days 1 to 4, and zuranolone 60 mg was administered on Day 5.

32) Standard deviation of lateral position when driving 100 km at the speed limit of 95 km/hour, maintaining a steady position in their lane, within the simulator.

33) The difference from placebo for the pooled mean of the multiple-dose zuranolone 30 mg and zuranolone 30/60 mg groups on Day 2.

3.59 [1.81, 5.37] cm, respectively. The placebo-corrected mean SDLPs in the morning on Day 6 after 5 days of dosing of zuranolone 30 mg, zuranolone 30/60 mg, or zopiclone [95% CI] were 1.07 [−0.52, 2.65], 6.48 [4.88, 8.08], and 1.73 [0.21, 3.34] cm, respectively. The upper bound of the 95% CI for the placebo-corrected SDLP after a single dose of zuranolone 30 mg or after 5 days of dosing of zuranolone 30/60 mg exceeded the pre-specified threshold of 4.4 cm.³⁴⁾

6.2.5.4 Interaction study with alprazolam or ethanol (CTD 5.3.3.4-03, Study 217-CLP-111 [■] to [■] 20[■])

Non-Japanese healthy adult subjects (25 subjects in Part A and 24 subjects in Part B included in pharmacodynamic assessment) received placebo or zuranolone 30 mg orally once daily after breakfast for 6 days in a crossover fashion, and placebo or zuranolone 30 mg was coadministered with alprazolam 1 mg (Part A) or ethanol (0.7 g/kg in male subjects, 0.6 g/kg in female subjects) (Part B) on Day 6. Cognitive function and alertness were assessed. Coadministration of zuranolone 30 mg with alprazolam or ethanol was associated with cognitive impairment (declines in visual learning and executive function) compared with 5 days of dosing of zuranolone alone, but the degree of impairment was similar to that with alprazolam alone or ethanol alone, and there was no trend towards greater sedation.

6.2.6 PPK analysis (CTD 5.3.3.5-01, 5.3.3.5-02)

Using the plasma pharmacokinetic data of zuranolone obtained from 18 Japanese and foreign clinical studies³⁵⁾ (1577 subjects, a total of 13744 PK samples), a population pharmacokinetics (PPK) analysis was performed (NONMEM version 7.4). The pharmacokinetics of zuranolone after oral administration were described by a 2-compartment model with a double transit compartment absorption model and first-order elimination. Based on the results of covariate exploration,³⁶⁾ body weight was selected as a statistically significant covariate on clearance (CL/F), and sex and body weight were selected as statistically significant covariates on apparent volume of distribution in central compartment (Vc/F) and apparent volume of distribution in peripheral compartment (Vp/F).

The effects of sex and body weight, the covariates in the final model, on the plasma pharmacokinetic parameters of zuranolone (C_{max} , C_{trough} , AUC) following repeated oral administration of zuranolone 30 mg once daily for 14 days were evaluated. The median ratios of the pharmacokinetic parameters in female patients vs. male patients were all within the range of 0.8 to 1.25. The median ratios of the pharmacokinetic parameters in patients with the median body weight vs. patients at the 95th percentile of body weight or at the 5th percentile of body weight were all within the range of 0.8 to 1.25.

Based on this PPK model, the C_{max} and AUC_{0-24h} on Day 14 following oral administration of zuranolone 30 mg

34) The threshold was chosen based on SDLP difference from placebo at 0.05% blood alcohol concentration obtained using a driving simulator.

35) Eleven Japanese and foreign clinical studies in healthy adult subjects (Studies 217-CLP-103, 217-CLP-106, 217-CLP-107, 217-CLP-108, 217-CLP-109, 217-CLP-110, 217-CLP-111, 217-CLP-114, 217-CLP-115, 217-EMX-101, and A3711), 5 Japanese and foreign clinical studies in patients with MDD (Studies 217-MDD-201B, 217-MDD-301A, 217-MDD-301B, A3731, and A3734), and 2 foreign clinical studies in patients with postpartum depression (Studies 217-PPD-201B and 217-PPD-301)

36) Covariates tested: sex, race (Caucasian/Black or African American/Asian/Others), body weight, age, eGFR, hepatic impairment (normal/mild impairment/moderate impairment/severe impairment)

once daily for 14 days in patients with depressive symptoms in Japanese and foreign clinical studies (Studies A3731, A3734, 217-MDD-201B,³⁷⁾ 217-MDD-301A,³⁸⁾ and 217-PPD-201B³⁹⁾) were estimated using Bayesian methods. The median $C_{\max}$ and AUC_{0-24h} were estimated at 67.2 ng/mL and 943.7 ng·h/mL, respectively.

6.R Outline of the review conducted by PMDA

6.R.1 Food effect on pharmacokinetics and the timing of dosing relative to meals

The applicant's explanation about the effect of food on the pharmacokinetics of orally administered zuranolone and the timing of dosing relative to meals in the DOSAGE AND ADMINISTRATION section of the proposed package insert:

In a phase I study in Japanese healthy adult subjects (Part B of Study A3711), a high-fat meal increased the $C_{\max}$ and AUC of zuranolone (Table 20), possibly because lipids in food and bile secretion following the ingestion of food increased the solubilization and absorption of highly lipid-soluble zuranolone. Zuranolone was to be taken after a meal in a Japanese phase III study (Study A3734) etc., which demonstrated the efficacy and safety of zuranolone. Thus, the DOSAGE AND ADMINISTRATION section should specify that zuranolone should be taken after a meal.

PMDA's view:

A high-fat meal increased the $C_{\max}$ and AUC of zuranolone, as compared to fasted conditions, and Study A3734 etc. in which zuranolone was to be taken after a meal, demonstrated the efficacy and safety of zuranolone [see Sections 7.R.2 and 7.R.3]. Given these findings, the DOSAGE AND ADMINISTRATION section specifying that zuranolone should be taken after a meal, is appropriate.

6.R.2 Need for dose adjustment in patients with factors associated with changes in zuranolone exposure

6.R.2.1 Use of zuranolone in patients with renal or hepatic impairment

The applicant's explanation about the need for dose adjustment and a precautionary statement regarding the use of zuranolone in patients with renal or hepatic impairment:

In a foreign phase I study in subjects with normal renal function or renal impairment (Study 217-CLP-107), the AUC of zuranolone increased by approximately 1.3- to 1.5-fold in subjects with mild, moderate, or severe renal impairment, as compared to subjects with normal renal function (Table 23), but no serious adverse events were reported, and there were no clinically relevant safety concerns.

In a foreign phase I study in subjects with normal hepatic function or hepatic impairment (Study 217-CLP-108), no appreciable changes in the AUC of zuranolone were observed in subjects with mild

37) A placebo-controlled, randomized, double-blind, parallel-group study in patients with MDD aged 18 to 65 years (Reference data CTD 5.3.5.1-04). Placebo or zuranolone 30 mg (a dose reduction to 20 mg was allowed in case of tolerability issues) was to be administered orally once daily after the evening meal for 14 days.

38) A placebo-controlled, randomized, double-blind, parallel-group study in patients with MDD aged 18 to 65 years (Reference data CTD 5.3.5.1-05). Placebo or zuranolone 20 or 30 mg was to be administered orally once daily after the evening meal for 14 days.

39) A placebo-controlled, randomized, double-blind, parallel-group study in patients with postpartum depression aged 18 to 45 years (Reference data CTD 5.3.5.1-08). Placebo or zuranolone 30 mg (a dose reduction to 20 mg was allowed in case of tolerability issues) was to be administered orally once daily after the evening meal for 14 days.

or moderate hepatic impairment, as compared to subjects with normal hepatic function, and the AUC of zuranolone increased by approximately 1.6-fold in subjects with severe hepatic impairment (Table 24). However, no serious adverse events were reported in the study, and there were no clinically relevant safety concerns.

Given the following considerations in addition to the above study results, the package insert should advise that zuranolone exposure is increased in patients with renal impairment or severe hepatic impairment, but no dose adjustment is necessary.

- Among patients with renal impairment treated with zuranolone 30 mg (175 patients with mild renal impairment, 13 patients with moderate renal impairment, 2 patients with severe renal impairment) in a Japanese phase III study (1) (Study A3734), 1 patient with moderate renal impairment experienced serious adverse events (COVID-19 and liver function test abnormal) during the treatment period. A causal relationship to zuranolone could not be ruled out for liver function test abnormal, and the event led to treatment discontinuation, but had an outcome of resolved. The event may have been associated with heavy drinking on the previous day [see Section 7.R.3.1]. There were no clinically relevant safety concerns in patients with renal impairment. No patients with severe hepatic impairment were enrolled in the study.
- In a Japanese phase III study (2) (Study A3736), no serious adverse events occurred in patients with renal impairment treated with zuranolone 30 mg (47 patients with mild renal impairment, 2 patients with moderate renal impairment), and there were no clinically relevant safety concerns in patients with renal impairment. No patients with severe hepatic impairment were enrolled in the study.
- Zuranolone 50 mg was to be administered orally once daily after the evening meal for 14 days in a foreign phase III study in patients with MDD (Study 217-MDD-301B⁴⁰⁾). The PPK model [see Section 6.2.6]-predicted median AUC_{0-24h} on Day 14 at this dosing regimen was 1529 ng·h/mL, which was 1.62-fold the AUC_{0-24h} (943.7 ng·h/mL) at the clinical dose (30 mg/day) in Japan. Following administration of zuranolone 50 mg in this study, the incidence of adverse events was 55.2% (148 of 268 subjects). Serious adverse events occurred in 1 subject (psychotic disorder and slow speech), and a causal relationship to zuranolone could not be ruled out for both events, but these events had an outcome of resolved. There were no clinically relevant safety concerns about administration of zuranolone 50 mg compared with 30 mg.
- According to the approved dosage and administration in the US,⁴¹⁾ the recommended dose of zuranolone in patients with moderate or severe renal impairment and patients with severe hepatic impairment is 30 mg. According to the post-marketing safety information, no particular problems have been identified.

6.R.2.2 Concomitant use with CYP3A inhibitors or inducers

The applicant's explanation about concomitant use of zuranolone with CYP3A4 inhibitors or inducers:

40) A placebo-controlled, randomized, double-blind, parallel-group study in patients with MDD aged 18 to 64 years (Reference data CTD 5.3.5.1-06). Placebo or zuranolone 50 mg (a dose reduction to 40 mg was allowed in case of tolerability issues) was to be administered orally once daily after the evening meal for 14 days. Antidepressant use was permitted, if patients were on a stable dose for ≥60 days prior to Day 1 and remained on it until Day 42.

41) In the US, the usual dosage of zuranolone 50 mg taken orally once daily after the evening meal for 14 days has been approved.

As to concomitant use of zuranolone with CYP3A4 inhibitors, coadministration with a strong CYP3A inhibitor, itraconazole, increased the AUC of zuranolone by approximately 1.6-fold (Table 25) in a clinical drug interaction study, and concomitant use of strong CYP3A inhibitors was prohibited in Japanese phase III studies etc. However, the AUC of zuranolone following oral administration of zuranolone 30 mg (the clinical dose in Japan) with a strong CYP3A inhibitor is considered to be similar to the AUC of zuranolone following oral administration of zuranolone 50 mg alone. There were no clinically relevant safety concerns in a foreign phase III study in which zuranolone 50 mg was to be administered [see Section 6.R.2.1]. In the US, the recommended dose of zuranolone when used concomitantly with a strong CYP3A4 inhibitor is 30 mg,⁴¹⁾ and according to the post-marketing safety information, no particular problems have been identified. Given the above points etc., the Precautions for Co-administration section of the package insert should advise that coadministration with a strong CYP3A inhibitor increases zuranolone exposure, but no dosage modification is necessary when zuranolone is used concomitantly with a strong CYP3A inhibitor.

As to concomitant use of zuranolone with CYP3A4 inducers, since coadministration with a CYP3A inducer, rifampicin, decreased the AUC of zuranolone by approximately 0.15-fold in a clinical drug interaction study (Table 25), CYP3A inducers should be listed in the Precautions for Co-administration section of the package insert.

Based on Sections 6.R.2.1 and 6.R.2.2, PMDA's view on the need for dose adjustment in patients with factors associated with changes in zuranolone exposure:

The AUC of zuranolone tended to increase following administration of zuranolone in patients with renal impairment or severe hepatic impairment or following coadministration of zuranolone with a strong CYP3A inhibitor. Given the applicant's explanation in Sections 6.R.2.1 and 6.R.2.2, no dose adjustment is necessary when zuranolone is administered to patients with renal impairment or severe hepatic impairment and when zuranolone is administered concomitantly with a strong CYP3A inhibitor, and the package insert should advise about increases in zuranolone exposure. The patient's condition should be closely monitored following administration of zuranolone. Listing CYP3A inducers in the Precautions for Co-administration section is appropriate.

7. Clinical Efficacy and Safety and Outline of the Review Conducted by PMDA

The applicant submitted the main efficacy and safety evaluation data, in the form of the results from clinical studies presented in Table 28. The main study results are described below.

Table 28. Listing of main efficacy and safety clinical studies

Data category	Geographical location	Study ID CTD	Phase	Study population	Number of subjects	Dosing regimen	Main endpoints
Evaluation	Japan	A3711 5.3.3.1-01	I	Healthy adult subjects	Part A: 72 Part B: 12 Part C: 8	Part A: a single oral dose of placebo or zuranolone 10, 20, or 30 mg followed by 7 days of oral dosing Part B: a single oral dose of zuranolone 30 mg Part C: a single oral dose of placebo or zuranolone 10 or 30 mg	Safety Pharmacokinetics
	Japan	A3731 5.3.5.1-01	II	Patients with MDD	250 ^{a)}	Placebo or zuranolone 20 or 30 mg taken orally once daily for 14 days	Efficacy Safety
	Japan	A3734 5.3.5.1-02	III	Patients with MDD	Part A: 412 ^{a)} Part B: 307	Part A: Placebo or zuranolone 30 mg taken orally once daily for 14 days Part B: Zuranolone 30 mg taken orally once daily for 14 days, followed by ≥6 weeks off-treatment, repeated over 52 weeks.	Efficacy Safety
	Japan	A3736 5.3.5.1-03	III	Patients with MDD	Part A: 108 ^{a)} Part B: 82	Part A: Placebo or zuranolone 30 mg taken orally once daily for 14 days Part B: Zuranolone 30 mg taken orally once daily for 14 days, followed by ≥6 weeks off-treatment, repeated over 52 weeks.	Efficacy Safety

a) Number of randomized subjects

7.1 Phase I studies

7.1.1 Japanese phase I study (CTD 5.3.3.1-01, Study A3711 [■ 20■ to ■ 20■])

A clinical study was conducted to evaluate the safety etc. of single or multiple oral doses of zuranolone in healthy adult subjects. The study consisted of Part A (a single- and multiple-dose study), Part B (a food effect study [see Section 6.1.1]), and Part C (an electroencephalography study). Part A is described below.

In Part A, a placebo-controlled, double-blind, parallel-group study was conducted in Japanese and non-Japanese (Caucasian) healthy adult subjects (target sample size, 36 non-elderly Japanese subjects, 24 non-elderly non-Japanese subjects, 12 Japanese elderly subjects).

Subjects were to receive a single oral dose of placebo or zuranolone after breakfast followed 3 days later by placebo or zuranolone 10, 20, or 30 mg orally once daily after the evening meal for 7 days. The doses of zuranolone for non-elderly Japanese, non-elderly non-Japanese, and Japanese elderly subjects are shown in Table 29. Subjects in each treatment group were randomized in a 1:3 ratio to receive placebo or zuranolone.

Table 29. Zuranolone doses for non-elderly Japanese, non-elderly non-Japanese, and Japanese elderly subjects

	Non-elderly Japanese subjects			Non-elderly non-Japanese subjects		Japanese elderly subjects
Zuranolone dose	10 mg	20 mg	30 mg	20 mg	30 mg	30 mg ^{a)}

a) Based on the results in non-elderly Japanese subjects, the maximum tolerated dose of 30 mg was selected. If the tolerability of zuranolone 30 mg in Japanese elderly subjects was not demonstrated, the zuranolone 10 or 20 mg group was planned to be added. Since there were no concerns about zuranolone 30 mg, the zuranolone 10 or 20 mg group was not added.

All of 72 randomized subjects were included in the safety population. One placebo-treated subject discontinued the study for a personal reason.

Adverse events occurred in 1 of 9 subjects in the placebo group, 1 of 9 subjects in the zuranolone 10 mg group, and 2 of 9 subjects in the zuranolone 30 mg group among non-elderly Japanese subjects, 2 of 6 subjects in the placebo group, 4 of 9 subjects in the zuranolone 20 mg group, and 4 of 9 subjects in the zuranolone 30 mg

group among non-elderly non-Japanese subjects, and 1 of 9 subjects in the zuranolone 30 mg group among Japanese elderly subjects. There were no serious adverse events including death or adverse events leading to treatment discontinuation.

Adverse events reported by ≥ 2 subjects in any group were somnolence (1 non-elderly Japanese subject in the placebo group, 1 non-elderly Japanese subject in the zuranolone 30 mg group, 2 non-elderly non-Japanese subjects in the placebo group, 3 non-elderly non-Japanese subjects in the zuranolone 20 mg group, 3 non-elderly non-Japanese subjects in the zuranolone 30 mg group, 1 Japanese elderly subject in the zuranolone 30 mg group).

7.2 Phase II studies

7.2.1 Japanese phase II study (CTD 5.3.5.1-01, Study A3731 [June 2020 to September 2021])

A placebo-controlled, randomized, double-blind, parallel-group study was conducted to evaluate the efficacy and safety of zuranolone in patients diagnosed with MDD according to the Diagnostic and Statistical Manual of Mental Disorders Fifth Edition (DSM-5), aged 18 to 75 years (target sample size, 240 subjects⁴²⁾ [80 in the placebo group, 80 in the zuranolone 20 mg group, 80 in the zuranolone 30 mg group]).

The key inclusion criteria were as follows:

The current depressive episode was ongoing for ≥ 8 weeks but no more than 12 months prior to signing the informed consent form. A 17-item Hamilton rating scale for depression (HAM-D17) total score⁴³⁾ of ≥ 22 and a Patient Health Questionnaire-9 (PHQ-9) score⁴⁴⁾ of ≥ 15 .

The study consisted of a treatment period (2 weeks), a follow-up period A (6 weeks), and a follow-up period B (6 weeks).

Placebo or zuranolone 20 or 30 mg was to be administered orally once daily after the evening meal for 14 days.

Among 250 randomized subjects⁴⁵⁾ (83 in the placebo group, 85 in the zuranolone 20 mg group, 82 in the zuranolone 30 mg group), 242 subjects (79 in the placebo group, 84 in the zuranolone 20 mg group, 79 in the zuranolone 30 mg group) after excluding 1 subject who did not receive study drug (the placebo group) and 7 subjects with GCP deviations (3 in the placebo group, 1 in the zuranolone 20 mg group, 3 in the zuranolone 30 mg group) were included in the safety population and in the full analysis set (FAS). The FAS was used as the efficacy population. There were 10 discontinuations (3 in the placebo group, 4 in the zuranolone 20 mg group, 3 in the zuranolone 30 mg group) during the treatment period, 24 discontinuations (7 in the placebo

42) Assuming a treatment difference of -3.5 between zuranolone and placebo for the primary endpoint, a standard deviation of 7.5 , a two-sided significance level of 5% , a 80% power, and a dropout rate of 5% , a target sample size of 80 subjects per group was chosen.

43) The HAM-D-17 is a 17-item clinician-administered depression assessment scale. It contains 17 items pertaining to symptoms of depression, each scored on a 3- to 5-point scale from 0 to 2 , 0 to 3 , or 0 to 4 (0 represents "absent," and the highest score represents the most severe symptom). The total score is calculated by summing the points from all questions.

44) The PHQ-9 consists of 9 questions about symptoms of depression over the last 2 weeks, with each item scored by patient on a 4-point scale: 0 (Not at all), 1 (Several days), 2 (More than half the days), and 3 (Nearly every day). The total score is calculated by summing the points from all questions.

45) Subjects were randomized in a $1:1:1$ ratio to receive placebo, zuranolone 20 mg, or zuranolone 30 mg. The randomization was stratified by baseline HAM-D17 total score (<25 , ≥ 25) and sex.

group, 10 in the zuranolone 20 mg group, 7 in the zuranolone 30 mg group) during the follow-up period A, and 16 discontinuations⁴⁶⁾ (6 in the placebo group, 6 in the zuranolone 20 mg group, 4 in the zuranolone 30 mg group) during the follow-up period B. The main reason for discontinuations during the treatment period was withdrawal by subject (7 subjects [2 in the placebo group, 3 in the zuranolone 20 mg group, 2 in the zuranolone 30 mg group]). The main reasons for discontinuations during the follow-up period were withdrawal by subject (11 subjects during the follow-up period A [1 in the placebo group, 5 in the zuranolone 20 mg group, 5 in the zuranolone 30 mg group], 9 subjects during the follow-up period B [4 in the placebo group, 3 in the zuranolone 20 mg group, 2 in the zuranolone 30 mg group]) and lack of efficacy (8 subjects during the follow-up period A [4 in the placebo group, 3 in the zuranolone 20 mg group, 1 in the zuranolone 30 mg group], 6 subjects during the follow-up period B [2 in the placebo group, 2 in the zuranolone 20 mg group, 2 in the zuranolone 30 mg group]).

The primary endpoint of the change from baseline in the HAM-D17 total score at Day 15 (the day after the end of study treatment) is shown in Table 30. There were statistically significant differences between both doses of zuranolone (20 mg and 30 mg) and placebo.

Table 30. Change from baseline in HAM-D17 total score at Day 15 (FAS)

Treatment group	HAM-D17 total score ^{a)}		Change from baseline ^{b) c)}	vs. placebo ^{c)}	
	Baseline	Day 15		Treatment difference [95% CI]	P-value ^{d)}
Placebo	24.4 ± 2.0 (79)	18.3 ± 5.5 (79)	-6.1 ± 0.64		
Zuranolone 20 mg	24.9 ± 2.4 (84)	16.5 ± 6.1 (80)	-8.2 ± 0.63	-2.1 [-3.9, -0.3]	0.0204
Zuranolone 30 mg	24.7 ± 2.2 (79)	16.2 ± 6.2 (77)	-8.3 ± 0.65	-2.2 [-4.0, -0.5]	0.0146

a) Mean ± SD (Number of evaluable subjects)

b) LS mean ± SE

c) Calculated using a mixed-effects model repeated measures (MMRM) analysis with treatment, time point, and treatment-by-time point interaction as fixed effects and baseline HAM-D17 total score and sex as covariates. An unstructured covariance structure was used, and the Kenward-Roger method was used to adjust degrees of freedom.

d) A two-sided significance level of 5%. A fixed-sequence testing procedure was used to adjust for multiplicity of comparisons between placebo and each dose of zuranolone. Zuranolone 30 mg was tested first against placebo, followed by zuranolone 20 mg.

Table 31 shows the change from baseline in the HAM-D17 total score over time during the treatment period and the follow-up period A (through 6 weeks after the end of study treatment [Day 57]).

Table 31. Change from baseline in HAM-D17 total score^{a)} over time up to Day 57 (FAS)

	Day 3	Day 8	Day 15 ^{b)}	Day 22	Day 43	Day 57
Placebo ^{c)}	-1.6 ± 0.37 (78)	-4.1 ± 0.53 (79)	-6.1 ± 0.64 (79)	-7.6 ± 0.71 (76)	-8.5 ± 0.82 (71)	-8.7 ± 0.85 (68)
Zuranolone 20 mg ^{d)}	-2.6 ± 0.36 (83)	-5.8 ± 0.51 (83)	-8.2 ± 0.63 (80)	-8.8 ± 0.70 (80)	-10.8 ± 0.81 (74)	-10.5 ± 0.84 (69)
	-1.0 [-2.0, 0.1]	-1.7 [-3.1, -0.2]	-2.1 [-3.9, -0.3]	-1.2 [-3.2, 0.8]	-2.3 [-4.5, 0.0]	-1.8 [-4.2, 0.5]
Zuranolone 30 mg ^{d)}	-2.7 ± 0.37 (79)	-6.3 ± 0.53 (78)	-8.3 ± 0.65 (77)	-9.1 ± 0.72 (74)	-10.2 ± 0.83 (69)	-10.2 ± 0.87 (68)
	-1.1 [-2.2, -0.1]	-2.1 [-3.6, -0.7]	-2.2 [-4.0, -0.5]	-1.5 [-3.5, 0.5]	-1.7 [-4.0, 0.6]	-1.5 [-3.9, 0.9]

a) Calculated using an MMRM analysis with treatment, time point, and treatment-by-time point interaction as fixed effects and baseline HAM-D17 total score and sex as covariates. An unstructured covariance structure was used, and the Kenward-Roger method was used to adjust degrees of freedom.

b) The primary time point (Study drug was not administered on Day 15 onwards.)

c) LS mean ± SE (Number of evaluable subjects)

d) Upper row, LS mean ± SE (Number of evaluable subjects); Lower row, Difference from placebo [95% CI]

Regarding safety, the incidences of all adverse events and adverse events reported by ≥2% of subjects in any group are shown in Table 32.

46) Including subjects who did not proceed from the follow-up period A to the follow-up period B.

Table 32. Incidences of all adverse events and adverse events reported by $\geq 2\%$ of subjects in any group (Safety population)

	Placebo	Zuranolone 20 mg	Zuranolone 30 mg
Number of evaluable subjects	79	84	79
All adverse events	41 (51.9)	46 (54.8)	43 (54.4)
Serious adverse events	0	0	0
Adverse events leading to treatment discontinuation	0	1 (1.2)	0
Adverse events reported by $\geq 2\%$ of subjects in any group			
Somnolence	5 (6.3)	10 (11.9)	17 (21.5)
Dizziness	3 (3.8)	9 (10.7)	7 (8.9)
Nasopharyngitis	4 (5.1)	6 (7.1)	6 (7.6)
Headache	3 (3.8)	6 (7.1)	4 (5.1)
Nausea	4 (5.1)	4 (4.8)	3 (3.8)
Cystitis	1 (1.3)	0	2 (2.5)
Seasonal allergy	0	0	2 (2.5)
Constipation	2 (2.5)	3 (3.6)	2 (2.5)
Diarrhoea	0	2 (2.4)	2 (2.5)
Blood creatine phosphokinase increased	1 (1.3)	1 (1.2)	2 (2.5)
Haemorrhoids	0	0	2 (2.5)
Feeling hot	0	0	2 (2.5)
Malaise	2 (2.5)	0	2 (2.5)
Thirst	1 (1.3)	0	2 (2.5)
Contusion	1 (1.3)	0	2 (2.5)
Urticaria	0	0	2 (2.5)
Abdominal discomfort	1 (1.3)	2 (2.4)	1 (1.3)
Urine leukocyte esterase positive	2 (2.5)	2 (2.4)	1 (1.3)
ALT increased	2 (2.5)	1 (1.2)	1 (1.3)
Blood uric acid increased	2 (2.5)	1 (1.2)	1 (1.3)
Restless legs syndrome	2 (2.5)	0	1 (1.3)
Abdominal pain upper	2 (2.5)	0	1 (1.3)
Back pain	2 (2.5)	3 (3.6)	0
Feeling abnormal	2 (2.5)	1 (1.2)	0
Nightmare	2 (2.5)	0	0

n (Incidence [%])

No serious adverse events including death were reported in any group. Adverse events leading to treatment discontinuation occurred in 1 subject in the zuranolone 20 mg group (dizziness and somnolence), and a causal relationship to study drug could not be ruled out for both events.

7.3 Phase III studies

7.3.1 Japanese phase III study (1) (CTD 5.3.5.1-02, Study A3734 [February 2022 to May 2024])

A placebo-controlled, randomized, double-blind, parallel-group study was conducted to evaluate the efficacy and safety of zuranolone in patients diagnosed with MDD according to the DSM-5 aged 18 to 75 years (target sample size, 400 subjects⁴⁷⁾ [200 in the placebo group, 200 in the zuranolone 30 mg group]).

The key inclusion criteria were as follows:

The current MDD episode was ongoing for ≥ 8 weeks but no more than 12 months prior to signing the informed consent form. A HAM-D17 total score of ≥ 22 and a PHQ-9 score of ≥ 15 .

The study consisted of a double-blind phase (Part A, 8 weeks) and an open-label extension phase (Part B, 52 weeks⁴⁸⁾). Subjects who completed the double-blind phase were allowed to enter the open-label extension phase if no safety issues were identified. The double-blind phase consisted of a treatment period (2 weeks) and a

47) Assuming a treatment difference of -2.1 between zuranolone 30 mg and placebo for the primary endpoint, a standard deviation of 6.1, a two-sided significance level of 5%, a 90% power, and a dropout rate of 10%, a target sample size of 200 subjects per group was chosen.

48) The open-label extension phase was 52 weeks from the first treatment with zuranolone during the open-label extension phase following the double-blind phase. For subjects who did not receive zuranolone during the open-label extension phase, the open-label extension phase was 52 weeks from study treatment during the double-blind phase.

follow-up period (6 weeks). The open-label extension phase consisted of a treatment period (2 weeks), a follow-up period (6 weeks), and a durability observation period (until a treatment decision was made after the follow-up period⁴⁹⁾), and one treatment cycle of zuranolone was defined as a treatment period and a follow-up period.

During the treatment period of the double-blind phase, placebo or zuranolone 30 mg was to be administered orally once daily after the evening meal for 14 days. During the treatment period of the open-label extension phase, zuranolone 30 mg was to be administered orally once daily after the evening meal for 14 days. If a subject met the criteria for starting treatment⁵⁰⁾ in Part B at least 6 weeks after the end of dosing (at the end of the follow-up period) during the double-blind phase and the open-label extension phase, they were to enter the treatment period⁵¹⁾ and receive treatment with zuranolone (a 14-day administration of zuranolone). During the open-label extension phase, the maximum number of treatment cycles was 6.

Among 412 randomized subjects⁵²⁾ in the double-blind phase (205 in the placebo group, 207 in the zuranolone 30 mg group), 404 subjects (199 in the placebo group, 205 in the zuranolone 30 mg group) after excluding 5 subjects with GCP deviations (4 in the placebo group, 1 in the zuranolone 30 mg group), 1 untreated subject with no HAM-D17 data (the placebo group), 1 subject with no HAM-D17 data (the zuranolone 30 mg group), and 1 untreated subject (the placebo group) were included in the safety population and in the FAS during the double-blind phase. The FAS was used as the efficacy population. There were 35 discontinuations during the double-blind phase (11 in the placebo group, 24 in the zuranolone 30 mg group), and the main reasons for discontinuations were lack of efficacy (10 subjects [2 in the placebo group, 8 in the zuranolone 30 mg group]) and withdrawal by subject (9 subjects [4 in the placebo group, 5 in the zuranolone 30 mg group]).

Among 377 subjects who completed the double-blind phase (194 in the placebo group, 183 in the zuranolone 30 mg group), 307 subjects (162 in the placebo group, 145 in the zuranolone 30 mg group) entered the open-label extension phase. After excluding 33 subjects who did not receive treatment with zuranolone during the open-label extension phase and 3 subjects with GCP deviations, 271 subjects were included in the safety and efficacy populations during the open-label extension phase. There were 174 discontinuations during the open-label extension phase (the number of zuranolone treatment cycles: 0 in 7 subjects, 1 in 45 subjects, 2 in 31 subjects, 3 in 10 subjects, 4 in 10 subjects, 5 in 9 subjects, 6 in 62 subjects), and the main reasons for discontinuations were withdrawal by subject (61 subjects) among subjects who received 0 to 5 treatment cycles and the re-treatment criteria met after the completion of treatment cycle 6 (61 subjects) among subjects who received 6 treatment cycles.

Table 33 shows the primary endpoint of the change from baseline in the HAM-D17 total score at Day 15 (the

49) After the follow-up period, a treatment decision was made every 8 weeks. If the PHQ-9 score was ≥ 10 , or if the physician suspected that treatment might be required, the subject returned to the study site for a treatment decision visit.

50) Subjects were considered to meet the criteria for starting treatment if all of the following criteria were met.

- A HAM-D17 total score of ≥ 14
- Depressive episodes (based on the DSM-5) persisting for ≥ 2 weeks

51) If a subject did not meet the criteria for starting treatment, they were to enter the durability observation period of the open-label extension phase and were not to enter the treatment period or receive zuranolone until the criteria for starting treatment were met.

52) Subjects were randomized in a 1:1 ratio to receive placebo or zuranolone 30 mg. The randomization was stratified by baseline HAM-D17 total score (<25 , ≥ 25), sex, and prior treatment for the current depressive episode (Yes, No).

day after the end of study treatment [the treatment period] during the double-blind phase). There was a statistically significant difference between the zuranolone 30 mg and placebo groups.

Table 33. Change from baseline in HAM-D17 total score at Day 15 (FAS)

Treatment group	Baseline	Day 15	Change from baseline ^{a) b)}	vs. placebo ^{b)}	
				Treatment difference in change [95% CI]	P-value ^{c)}
Placebo	24.1 ± 2.1 (199)	17.8 ± 5.6 (194)	-6.2 ± 0.41		
Zuranolone 30 mg	24.1 ± 2.0 (205)	16.7 ± 5.8 (192)	-7.4 ± 0.40	-1.2 [-2.3, -0.1]	0.0365

Mean ± SD (Number of evaluable subjects)

a) LS mean ± SE

b) Calculated using an MMRM analysis with treatment, time point, and treatment-by-time point interaction as fixed effects and baseline HAM-D17 total score, sex, and prior drug treatment for the current depressive episode (Yes, No) as covariates. An unstructured covariance structure was used, and the Kenward-Roger method was used to adjust degrees of freedom.

c) A two-sided significance level of 5%

Table 34 shows the change from baseline in the HAM-D17 total score over time during the double-blind phase.

Table 34. Change from baseline in HAM-D17 total score^{a)} over time up to Day 57 (FAS)

	Day 3	Day 8	Day 15 ^{b)}	Day 22	Day 43	Day 57
Placebo ^{c)}	-1.7 ± 0.23 (199)	-4.0 ± 0.32 (196)	-6.2 ± 0.41 (194)	-7.2 ± 0.41 (195)	-8.2 ± 0.49 (188)	-7.7 ± 0.50 (184)
Zuranolone 30 mg ^{d)}	-3.2 ± 0.23 (201)	-5.7 ± 0.32 (198)	-7.4 ± 0.40 (192)	-7.8 ± 0.41 (187)	-8.6 ± 0.49 (178)	-8.7 ± 0.50 (175)
	-1.5 [-2.2, -0.9]	-1.8 [-2.6, -0.9]	-1.2 [-2.3, -0.1]	-0.6 [-1.7, 0.6]	-0.5 [-1.8, 0.9]	-1.0 [-2.4, 0.4]

a) Calculated using an MMRM analysis with treatment, time point, and treatment-by-time point interaction as fixed effects and baseline HAM-D17 total score, sex, and prior drug treatment for the current depressive episode (Yes, No) as covariates. An unstructured covariance structure was used, and the Kenward-Roger method was used to adjust degrees of freedom.

b) The primary time point (Study drug was not administered on Day 15 onwards.)

c) LS mean ± SE (Number of evaluable subjects)

d) Upper row, LS mean ± SE (Number of evaluable subjects); Lower row, Difference from placebo [95% CI]

Table 35 shows the change from baseline (the last value prior to dosing in each cycle) in the HAM-D17 total score on the day after the end of dosing (the treatment period) (Day 15) in each treatment cycle of zuranolone during the open-label extension phase.

Table 35. Change from baseline in HAM-D17 total score on the day after the end of dosing (Day 15) in each treatment cycle of zuranolone during the open-label extension phase

Treatment group during the double-blind phase	Number of treatment cycles of zuranolone					
	Cycle 1	Cycle 2	Cycle 3	Cycle 4	Cycle 5	Cycle 6
Placebo	-4.8 ± 4.8 (126)	-5.4 ± 4.6 (104)	-5.0 ± 4.4 (89)	-4.5 ± 4.8 (83)	-4.8 ± 5.2 (63)	-4.5 ± 4.5 (54)
Zuranolone 30 mg	-5.3 ± 4.8 (121)	-4.9 ± 4.9 (96)	-4.1 ± 4.4 (74)	-4.4 ± 4.5 (60)	-4.2 ± 4.2 (52)	-3.6 ± 3.7 (41)

Mean ± SD (Number of evaluable subjects)

Regarding safety, Table 36 shows the incidences of all adverse events and adverse events reported by ≥2% of subjects in either group or during any period of the open-label extension phase.

Table 36. Incidences of all adverse events and adverse events reported by $\geq 2\%$ of subjects in either group or during any period of the open-label extension phase (Safety population)

	Double-blind phase				Open-label extension phase		
	Treatment period		Follow-up period		Treatment period	Follow-up period	Durability observation period
	Placebo	Zuranolone 30 mg	Placebo	Zuranolone 30 mg			
Number of evaluable subjects	199	205	199	205	304	304	201
All adverse events	45 (22.6)	96 (46.8)	48 (24.1)	50 (24.4)	159 (52.3)	131 (43.1)	50 (24.9)
Serious adverse events	0	0	0	0	3 (1.0)	4 (1.3)	0
Adverse events leading to treatment discontinuation	0	2 (1.0)	0	0	8 (2.6)	0	0
Adverse events reported by $\geq 2\%$ of subjects in either group							
Somnolence	11 (5.5)	27 (13.2)	1 (0.5)	0	45 (14.8)	1 (0.3)	0
Dizziness	2 (1.0)	25 (12.2)	1 (0.5)	1 (0.5)	29 (9.5)	0	2 (1.0)
Feeling abnormal	1 (0.5)	13 (6.3)	0	0	15 (4.9)	1 (0.3)	0
Headache	3 (1.5)	7 (3.4)	1 (0.5)	3 (1.5)	10 (3.3)	7 (2.3)	3 (1.5)
Nausea	2 (1.0)	7 (3.4)	0	1 (0.5)	7 (2.3)	7 (2.3)	1 (0.5)
COVID-19	3 (1.5)	5 (2.4)	6 (3.0)	3 (1.5)	8 (2.6)	18 (5.9)	10 (5.0)
Asthenia	0	4 (2.0)	0	1 (0.5)	6 (2.0)	0	0
Vertigo	0	4 (2.0)	0	0	2 (0.7)	1 (0.3)	0
Pyrexia	0	3 (1.5)	3 (1.5)	7 (3.4)	11 (3.6)	16 (5.3)	4 (2.0)
Diarrhoea	3 (1.5)	3 (1.5)	1 (0.5)	5 (2.4)	5 (1.6)	7 (2.3)	2 (1.0)
Nasopharyngitis	3 (1.5)	2 (1.0)	3 (1.5)	6 (2.9)	13 (4.3)	25 (8.2)	8 (4.0)
Myalgia	1 (0.5)	1 (0.5)	1 (0.5)	4 (2.0)	1 (0.3)	2 (0.7)	1 (0.5)
Back pain	0	1 (0.5)	1 (0.5)	1 (0.5)	7 (2.3)	7 (2.3)	2 (1.0)
ALT increased	1 (0.5)	1 (0.5)	2 (1.0)	0	5 (1.6)	6 (2.0)	1 (0.5)

n (Incidence [%])

No deaths were reported. Serious adverse events other than death occurred in 3 subjects during the treatment period of the open-label extension phase (altered state of consciousness; COVID-19 and liver function test abnormal; and mechanical ileus [1 subject each]) and 4 subjects during the follow-up period of the open-label extension phase (large intestine polyp and cerebral infarction; vertigo positional; pneumothorax spontaneous; and malignant pleural effusion and invasive ductal breast carcinoma [1 subject each]), and a causal relationship to study drug could not be ruled out for the events of altered state of consciousness and liver function test abnormal observed during the treatment period. Adverse events leading to treatment discontinuation occurred in 2 subjects in the zuranolone 30 mg group during the treatment period of the double-blind phase (dizziness; and dizziness, headache, nausea, and malaise [1 subject each]) and 8 subjects during the treatment period of the open-label extension phase (liver function test abnormal; feeling drunk; akathisia; palpitations and nausea; somnolence; headache, dizziness, and malaise; dizziness; and altered state of consciousness [1 subject each]), and a causal relationship to study drug could not be ruled out for all those events.

7.3.2 Japanese phase III study (2) (CTD 5.3.5.1-03, Study A3736 [November 2022 to March 2024])

A placebo-controlled, randomized, double-blind, parallel-group study was conducted to evaluate the efficacy and safety of zuranolone in patients diagnosed with MDD according to the DSM-5 aged 18 to 75 years who had been taking 1 antidepressant (SSRI, serotonin-noradrenaline reuptake inhibitor [SNRI], or vortioxetine hydrobromide) for ≥ 4 weeks (target sample size, 100 subjects⁵³⁾ [50 in the placebo group, 50 in the zuranolone 30 mg group]).

53) Assuming a HAM-D17 response rate in the placebo group of 20% and an odds ratio of 2.1 for the HAM-D17 response rate (the proportion of subjects with a $\geq 50\%$ reduction in HAM-D17 total score from baseline) for zuranolone 30 mg vs. placebo, 43 subjects per group were required to detect an odds ratio of >1.14 for the HAM-D17 response rate for zuranolone 30 mg vs. placebo with $\geq 90\%$ power. Then, a target sample size of 100 subjects (50 per group) was chosen.

The key inclusion criteria were as follows:

The current depressive episode was ongoing for ≥ 8 weeks but no more than 12 months prior to signing the informed consent form. A HAM-D17 total score of ≥ 14 and a PHQ-9 score of ≥ 10 .

The study consisted of a double-blind phase (Part A, 8 weeks) and an open-label extension phase (Part B, 52 weeks⁴⁸⁾). Subjects who completed the double-blind phase were allowed to enter the open-label extension phase if no safety issues were identified. The double-blind phase consisted of a treatment period (2 weeks) and a follow-up period (6 weeks). The open-label extension phase consisted of a treatment period (2 weeks), a follow-up period (6 weeks), and a durability observation period (until a treatment decision was made after the follow-up period⁴⁹⁾). One treatment cycle of zuranolone was defined as a treatment period and a follow-up period. Since the primary endpoint during the double-blind phase was not met (Table 37), the open-label extension phase was terminated early due to the sponsor's (the applicant's) decision.

During the treatment period of the double-blind phase, placebo or zuranolone 30 mg was to be administered orally once daily after the evening meal for 14 days. During the treatment period of the open-label extension phase, zuranolone 30 mg was to be administered orally once daily after the evening meal for 14 days. If a subject met the criteria for starting treatment⁵⁰⁾ in Part B at least 6 weeks after the end of dosing (at the end of the follow-up period) during the double-blind phase and the open-label extension phase, they were to enter the treatment period⁵¹⁾ and receive treatment with zuranolone (a 14-day administration of zuranolone). During the open-label extension phase, the maximum number of treatment cycles was 6. During the study period, subjects were to remain on a stable dosage of 1 antidepressant initiated prior to signing the informed consent form.

Among 108 randomized subjects⁵⁴⁾ in the double-blind phase (53 in the placebo group, 55 in the zuranolone 30 mg group), 107 subjects (52 in the placebo group, 55 in the zuranolone 30 mg group) after excluding 1 untreated subject (the placebo group) were included in the safety population and in the FAS. The FAS was used as the efficacy population. There were 7 discontinuations during the double-blind phase (4 in the placebo group, 3 in the zuranolone 30 mg group), and the main reason for discontinuations was withdrawal by subject (3 subjects [2 in the placebo group, 1 in the zuranolone 30 mg group]).

Among 101 subjects who completed the double-blind phase (49 in the placebo group, 52 in the zuranolone 30 mg group), 82 subjects (35 in the placebo group, 47 in the zuranolone 30 mg group) entered the open-label extension phase. After excluding 6 subjects who did not receive treatment with zuranolone during the open-label extension phase, 76 subjects were included in the safety and efficacy populations during the open-label extension phase. There were 77 discontinuations during the open-label extension phase (the number of zuranolone treatment cycles, 0 in 5 subjects, 1 in 15 subjects, 2 in 16 subjects, 3 in 11 subjects, 4 in 13 subjects, 5 in 6 subjects, 6 in 11 subjects), including 48 discontinuations due to the sponsor decision. Other main reasons for discontinuations were withdrawal by subject (11 subjects) among subjects who received 0 to 5 treatment cycles and the re-treatment criteria met after the completion of treatment cycle 6 (6 subjects) among

54) Subjects were randomized in a 1:1 ratio to receive placebo or zuranolone 30 mg. The randomization was stratified by baseline HAM-D17 total score (<20 , ≥ 20) and sex.

subjects who received 6 treatment cycles.

Table 37 shows the primary endpoint of the HAM-D17 response rate⁵⁵⁾ at Day 15 (the day after the end of study treatment [the treatment period] during the double-blind phase). The point estimate of the adjusted odds ratio for zuranolone 30 mg vs. placebo was 0.56, which was below the pre-specified threshold of 1.14.

Table 37. HAM-D17 response rate at Day 15 during the double-blind phase (FAS)

Treatment group	HAM-D17 total score			HAM-D17 response rate ^{b)}	Odds ratio [95% CI] ^{c)}
	Baseline	Day 15	Change ^{a)}		
Placebo	20.8 ± 3.4 (52)	15.3 ± 5.6 (51)	-5.2 ± 0.74	21.6% (11/51)	0.56 [0.20, 1.59]
Zuranolone 30 mg	20.1 ± 3.0 (55)	15.3 ± 5.0 (53)	-4.9 ± 0.72	13.2% (7/53)	

Mean ± SD (Number of evaluable subjects)

a) LS mean ± SE. Calculated using an MMRM analysis with treatment, time point, and treatment-by-time point interaction as fixed effects and baseline HAM-D17 total score and sex as covariates. An unstructured covariance structure was used, and the Kenward-Roger method was used to adjust degrees of freedom.

b) Proportion (Number of subjects)

c) Inverse probability-weighted generalized estimating equation (IPW-GEE) with treatment, time point, and treatment-by-time point interaction as fixed effects and baseline HAM-D17 total score and sex as covariates. IPW-GEE assumed an independent structure for the working correlation matrix.

Table 38 shows the change from baseline in the HAM-D17 total score over time during the double-blind phase.

Table 38. Change from baseline in HAM-D17 total score^{a)} over time up to Day 57 (FAS)

	Day 3	Day 8	Day 15 ^{b)}	Day 22	Day 43	Day 57
Placebo ^{c)}	-2.1 ± 0.48 (52)	-3.8 ± 0.64 (52)	-5.2 ± 0.74 (51)	-5.6 ± 0.74 (50)	-5.9 ± 0.73 (49)	-6.3 ± 0.84 (46)
Zuranolone 30 mg ^{d)}	-1.5 ± 0.47 (55)	-3.6 ± 0.62 (55)	-4.9 ± 0.72 (53)	-3.9 ± 0.71 (52)	-4.7 ± 0.71 (52)	-4.2 ± 0.81 (52)
	0.6 [-0.7, 2.0]	0.3 [-1.5, 2.0]	0.3 [-1.8, 2.4]	1.7 [-0.4, 3.7]	1.2 [-0.8, 3.2]	2.0 [-0.3, 4.4]

a) Calculated using an MMRM analysis with treatment, time point, and treatment-by-time point interaction as fixed effects and baseline HAM-D17 total score and sex as covariates. An unstructured covariance structure was used, and the Kenward-Roger method was used to adjust degrees of freedom.

b) The primary time point (Study drug was not administered on Day 15 onwards.)

c) LS mean ± SE (Number of evaluable subjects)

d) Upper row, LS mean ± SE (Number of evaluable subjects); Lower row, Difference from placebo [95% CI]

Regarding safety, Table 39 shows the incidences of all adverse events and adverse events reported by ≥3 subjects in either group or during any period of the open-label extension phase.

55) Proportion of subjects with a ≥50% reduction in HAM-D17 total score from baseline

Table 39. Incidences of all adverse events and adverse events reported by ≥ 3 subjects in either group or during any period of the open-label extension phase (Safety population)

	Double-blind phase				Open-label extension phase		
	Treatment period		Follow-up period		Treatment period	Follow-up period	Durability observation period
	Placebo	Zuranolone 30 mg	Placebo	Zuranolone 30 mg			
Number of evaluable subjects	52	55	52	55	82	82	46
All adverse events	12 (23.1)	27 (49.1)	15 (28.8)	20 (36.4)	41 (50.0)	42 (51.2)	16 (34.8)
Serious adverse events	1 (1.9)	0	0	0	1 (1.2)	0	0
Adverse events leading to treatment discontinuation	0	1 (1.8)	0	0	4 (4.9)	0	0
Adverse events reported by ≥ 3 subjects in either group							
Somnolence	2 (3.8)	11 (20.0)	0	0	10 (12.2)	0	0
Dizziness	1 (1.9)	4 (7.3)	0	0	5 (6.1)	2 (2.4)	1 (2.2)
Nausea	0	4 (7.3)	2 (3.8)	3 (5.5)	3 (3.7)	1 (1.2)	0
Nasopharyngitis	0	2 (3.6)	2 (3.8)	3 (5.5)	5 (6.1)	16 (19.5)	8 (17.4)
Thirst	1 (1.9)	2 (3.6)	0	0	3 (3.7)	0	0
Sinusitis	0	1 (1.8)	0	0	0	3 (3.7)	0
COVID-19	0	0	1 (1.9)	0	2 (2.4)	5 (6.1)	1 (2.2)
Influenza	0	0	0	1 (1.8)	0	5 (6.1)	1 (2.2)
Headache	1 (1.9)	0	0	1 (1.8)	0	4 (4.9)	1 (2.2)
Vomiting	0	0	1 (1.9)	0	1 (1.2)	3 (3.7)	0
Back pain	1 (1.9)	0	3 (5.8)	0	1 (1.2)	2 (2.4)	0

n (Incidence [%])

No deaths were reported. Serious adverse events other than death occurred in 1 subject in the placebo group during the treatment period of the double-blind phase (hepatic function abnormal) and 1 subject during the treatment period of the open-label extension phase (hepatic function abnormal), and a causal relationship to study drug could not be ruled out for the event of hepatic function abnormal observed during the treatment period of the double-blind phase. Adverse events leading to treatment discontinuation occurred in 1 subject in the zuranolone 30 mg group during the treatment period of the double-blind phase (asthenia) and 4 subjects during the treatment period of the open-label extension phase (dizziness [2 subjects]; and insomnia; and hepatic function abnormal [1 subject each]), and a causal relationship to study drug could not be ruled out for all those events except for hepatic function abnormal.

7.R Outline of the review conducted by PMDA

7.R.1 Japanese phase III study plan and strategy for efficacy evaluation of zuranolone

The applicant's explanation about the Japanese phase III study plan for zuranolone:

Zuranolone binds at an allosteric site on GABA_A receptors and enhances GABA_A receptor function. Benzodiazepines with a different binding site enhance GABA_A receptor function in the same manner and are associated with the risk of dependence. According to the foreign clinical practice guidelines, the recommended duration of use of benzodiazepines is approximately 2 weeks (Guidance on appropriate prescribing of benzodiazepines and z-drugs (BZRA) for the treatment of anxiety and insomnia. Health Research Board; 2021). Based on the above, it was considered that long-term continuous use of zuranolone should also be avoided. In the clinical development of zuranolone by Sage Therapeutics in the US, the duration of use of zuranolone was limited to 14 days, and a foreign phase II study in patients with MDD (Study 217-MDD-201B³⁷) etc. were conducted. Zuranolone demonstrated improvement in depressive symptoms at Day 15 (the day after the end of dosing) compared with placebo, and the effect of zuranolone tended to be sustained to a certain extent even after the end of dosing. Taking account of the status of development overseas etc., the duration of use was limited to 14 days also in Japan, and it was decided to develop zuranolone as an antidepressant, administered using intermittent courses (14 days of treatment followed by off-treatment and re-treatment

according to the patient's condition). Then, a Japanese phase II study in patients with MDD (Study A3731) was planned and conducted, which suggested the efficacy of zuranolone (Table 30).

As the typical pattern of treatment of depression with pharmacotherapy in Japan, there are 3 phases of treatment according to the treatment goals: the acute phase from treatment initiation to remission to improve symptoms, the continuation phase to prevent relapse after remission and aim at recovery, and the maintenance phase with continued treatment to prevent recurrence after recovery (Main points of diagnosis and treatment of depression-10 [in Japanese]. Japan Depression Center [the 4th edition]; 2017).

Given that zuranolone demonstrated rapid improvement in depressive symptoms by Day 15 (the day after the end of dosing), etc., in Study A3731 etc., and that chronic use of zuranolone should be avoided due to the risk of dependence, it was decided to plan Japanese phase III studies, on the premise that zuranolone is intended for acute phase treatment of patients diagnosed with a first episode of depression or patients with relapse/recurrence of depressive symptoms requiring treatment intervention.

It was decided to plan and conduct a Japanese phase III study (1) to confirm the efficacy of zuranolone monotherapy in the acute phase treatment of patients with MDD (Study A3734) [see Section 7.3.1]. Since in the case of other antidepressants, continued treatment is recommended to prevent relapse/recurrence also after acute phase treatment (the Japanese Guideline for Treatment of Depression) etc., zuranolone was expected to be used as an add-on to other antidepressants in clinical practice. Thus, it was also decided to plan and conduct a Japanese phase III study (2) to exploratorily evaluate the efficacy of zuranolone as an add-on to other antidepressants in patients with MDD (Study A3736).

At the time of planning Studies A3734 and A3736, a placebo-controlled, randomized, double-blind, parallel-group study to confirm the efficacy of zuranolone orally administered at 20 or 30 mg once daily after the evening meal for 14 days in patients with MDD (a foreign phase III study: Study 217-MDD-301A⁵⁶⁾) had already been conducted overseas. In this study, the primary endpoint of the change from baseline in the HAM-D17 total score at Day 15 (LS mean $\pm$ SE) was -11.1 ± 0.59 in the placebo group, -11.5 ± 0.62 in the zuranolone 20 mg group, and -12.5 ± 0.68 in the zuranolone 30 mg group, which showed no statistically significant differences between zuranolone 20 or 30 mg and placebo (treatment difference [95% CI] (*P*-value), $-0.4 [-2.0, 1.3]$ (*P* = 0.66) in the zuranolone 20 mg group, $-1.4 [-3.1, 0.3]$ (*P* = 0.12) in the zuranolone 30 mg group).⁵⁷⁾ Prior to planning and conducting Studies A3734 and A3736, the factors contributing to the above results were discussed as follows based on the different designs of the Japanese and foreign clinical studies,

56) A placebo-controlled, randomized, double-blind, parallel-group study in patients with MDD aged 18 to 65 years (Reference data CTD 5.3.5.1-05). Key inclusion criteria were patients with depressive symptoms persisting for ≥ 4 weeks, a MADRS (Montgomery Åsberg Depression Rating Scale) total score of ≥ 32 , and a HAM-D17 total score of ≥ 22 . Placebo or zuranolone 20 or 30 mg was to be administered orally once daily after the evening meal for 14 days. Concomitant antidepressant use was permitted, if patients were on a stable dose for ≥ 60 days prior to Day 1 and remained on it until Day 42. Five hundred seventy randomized subjects who received study drug (190 in the placebo group, 188 in the zuranolone 20 mg group, 192 in the zuranolone 30 mg group) were included in the safety population, and 482 subjects with a baseline HAM-D17 total score of ≥ 22 (157 in the placebo group, 159 in the zuranolone 20 mg group, 166 in the zuranolone 30 mg group) were included in the efficacy population.

57) The results in each group and treatment difference were calculated using an MMRM analysis with treatment, baseline HAM-D17 total score, antidepressant use at baseline, time point, and treatment-by-time point interaction as fixed effects. An unstructured covariance structure was used, and the Kenward-Roger method was used to adjust degrees of freedom. A two-sided significance level of 5% was used. If the comparison between the zuranolone 30 mg and placebo groups was significant, the comparison between the zuranolone 20 mg and placebo groups was planned to be tested.

etc.

- Concomitant use of other antidepressants was prohibited in Study A3731, but was permitted in Study 217-MDD-301A. Subgroup analyses of the primary endpoint of Study 217-MDD-301A according to concomitant antidepressant use were performed, and the results are shown in Table 40. While there was a trend towards improvement in the zuranolone group compared with the placebo group among the subgroup without concomitant antidepressant use, there was no trend towards improvement in the zuranolone group compared with the placebo group among the subgroup with concomitant antidepressant use.

Table 40. Change from baseline in HAM-D17 total score at Day 15 with or without concomitant antidepressant use (Study 217-MDD-301A, mFAS^{a)})

Concomitant antidepressant use	Treatment group	Number of evaluable subjects	Change from baseline at Day 15 ^{b) c)}	Treatment difference [95% CI] ^{c)}
No	Placebo	96	-9.9 ± 0.72	
	Zuranolone 20 mg	107	-11.0 ± 0.75	-1.1 [-3.1, 1.0]
	Zuranolone 30 mg	109	-12.5 ± 0.72	-2.5 [-4.5, -0.5]
Yes	Placebo	45	-13.4 ± 0.96	
	Zuranolone 20 mg	45	-12.3 ± 1.07	1.1 [-1.8, 3.9]
	Zuranolone 30 mg	44	-12.2 ± 1.43	1.2 [-2.3, 4.6]

a) Randomized subjects with a baseline HAM-D17 total score of ≥22 and at least 1 post-baseline observation

b) LS mean ± SE

c) Analysis based on an analysis model similar to the primary analysis model

- The subgroup of patients on concomitant antidepressants during the study period had an inadequate response to antidepressants used prior to enrollment. A certain number of patients with treatment-resistant clinical symptoms were enrolled in Study 217-MDD-301A, and this factor etc. may have affected the efficacy evaluation of zuranolone.

Outside Japan, based on the results from Study 217-MDD-301A etc., subsequent clinical studies were conducted, including a higher dose of zuranolone 50 mg. However, taking account of the factors contributing to failure to demonstrate the efficacy of zuranolone in the above-mentioned Study 217-MDD-301A and the results of Study A3731 suggesting the efficacy of zuranolone, etc., planning and conduct of new studies, i.e., Studies A3734 and A3736, based on the results from Study A3731, was considered appropriate.

PMDA asked the applicant to explain the appropriateness of the primary endpoints of Studies A3734 and A3736 and how to evaluate the efficacy of zuranolone based on the results from the 2 Japanese phase III studies.

The applicant's explanation:

In accordance with the guidelines for the clinical evaluation of antidepressants in Japan (PFSB/ELD Notification No. 1116-1 dated November 16, 2010), the same primary endpoint as Study A3731, i.e., the change from baseline in the HAM-D17 total score was chosen for Study A3734. As to the timing of the primary endpoint, in the case of the currently approved antidepressants such as SSRIs, 6 to 8 weeks of treatment are required to assess patient response, and Weeks 6 to 8 are selected as the primary time point also in clinical studies. However, Study A3731 showed statistically significant differences between the zuranolone and placebo groups for the primary endpoint of the change from baseline in the HAM-D17 total score at Day 15 (the day after the end of study treatment) (Table 30). Moreover, the difference in the change from baseline

between the zuranolone and placebo groups peaked at Day 15 (the day after the end of study treatment), and there was a trend towards improvements in the HAM-D17 total score in the zuranolone group compared with the placebo group up to 6 weeks after the end of study treatment (Day 57) (Table 31). Based on the above clinical study results etc., it was considered that the primary time point in Study A3734 to be planned and conducted to confirm the efficacy of zuranolone should be Day 15 (the day after the end of study treatment).

On the other hand, though choosing the same primary endpoint as Studies A3731 and A3734 for Study A3736 was considered, as the treatment effect of a drug as an add-on to other antidepressants is generally lower than that of monotherapy, a larger sample size than in Study A3734 was expected to be needed, and in terms of feasibility, it was considered difficult to conduct a statistical hypothesis test to confirm the efficacy of zuranolone as an add-on to other antidepressants. Thus, the following study design was selected because the study of this design can evaluate the efficacy of zuranolone as an add-on to other antidepressants to a certain extent and is feasible in Japan: The success criterion for the study was to be met if the study showed that the odds ratio of the HAM-D17 response rate⁵⁵⁾ at Day 15 (the day after the end of study treatment) for zuranolone as an add-on to other antidepressants was similar to that for zuranolone monotherapy. The primary endpoint of Study A3736 was to be met if the point estimate of the odds ratio of the HAM-D17 response rate at Day 15 for zuranolone vs. placebo was above the lower bound of the 95% CI for the odds ratio of the HAM-D17 response rate at Day 15 (the day after the end of study treatment) in Study A3731 (1.14). If this criterion was met, this would suggest that the efficacy of zuranolone as an add-on to other antidepressants is at least similar to that of zuranolone monotherapy. Thus, the primary endpoint chosen for Study A3736 was considered appropriate.

Based on the above designs of Studies A3734 and A3736, it was decided to evaluate the efficacy of zuranolone in patients with depression based mainly on the results from Study A3734 and exploratorily evaluate the usefulness of zuranolone as an add-on to other antidepressants based on the results from Study A3736.

PMDA's view:

Since zuranolone enhances GABA_A receptor function, and dependence studies indicated that zuranolone produces psychological and physical dependence [see Section 5.6.3], long-term continuous use of zuranolone is not appropriate due to the risk of dependence. Thus, it is understandable that the duration of use of zuranolone was limited to 14 days, and that zuranolone was developed as an antidepressant intended for acute phase treatment after marketing, based on the pharmacotherapy paradigm for patients with depression in Japan and the action profile of zuranolone. Outside Japan, Study 217-MDD-301A showed no statistically significant differences in the primary endpoint between zuranolone 20 mg or 30 mg and placebo. However, given the applicant's explanation about the factors contributing to these results and the results from the Japanese phase II study (Study A3731) etc., planning and conduct of Japanese phase III studies based on the results from Study A3731 is also understandable.

According to the Japanese Guideline for Treatment of Depression, antidepressants should be used as monotherapy, as a rule, and treatment should be continued for a certain period of time to prevent relapse/recurrence also after the remission of depressive symptoms. Taking account of the pharmacotherapy

paradigm for patients with depression in Japan, etc., zuranolone is also expected to be used with the currently approved antidepressants such as SSRIs after marketing, with a view to treatment to prevent relapse/recurrence after the remission of depressive symptoms. Thus, in addition to the Japanese phase III study (1) to confirm the efficacy of zuranolone monotherapy (Study A3734), the Japanese phase III study (2) to exploratorily evaluate the efficacy of zuranolone as an add-on to other antidepressants in patients with depression, to the extent possible, without a statistical hypothesis test of comparison between the placebo and zuranolone groups (Study A3736), were planned and conducted for efficacy evaluation of zuranolone. This is understandable.

The change from baseline in the HAM-D17 total score at Day 15 (the day after the end of study treatment) chosen as the primary endpoint for Study A3734 was appropriate, given the results from the Japanese phase II study (Study A3731) etc. In the treatment of patients with depression, the maintenance of effect is important to ensure that depressive symptoms do not worsen or relapse/recur also after acute phase treatment. Thus, when assessing the clinical meaningfulness of zuranolone, the efficacy of zuranolone, including the clinical course after the end of dosing, needs to be evaluated.

Although Study A3736 can evaluate the efficacy of zuranolone as an add-on to the currently approved antidepressants such as SSRIs to a certain extent, efficacy evaluation using the success criterion established from the standpoint of the feasibility of the study has limitations.

Based on the above, PMDA decided to evaluate the efficacy of zuranolone, focusing on the results from Study A3734, and discuss the efficacy of zuranolone as an add-on to the currently approved antidepressants such as SSRIs, to the extent possible, based on the results from Study A3736.

7.R.2 Efficacy

7.R.2.1 Efficacy of zuranolone

The applicant's explanation about the efficacy of zuranolone in patients with depression:

The results of the primary endpoint of Study A3734 demonstrated the superiority of zuranolone over placebo (Table 33). The trend towards improvement from baseline in the HAM-D17 total score in the zuranolone 30 mg group was sustained also at 6 weeks after the completion of the 14-day treatment course (Day 57), and the improvements tended to be greater in the zuranolone 30 mg group than in the placebo group (Table 34). Table 41 shows the results of secondary endpoints during the double-blind phase of Study A3734, i.e., the HAM-D17 response rate⁵⁵⁾ and the HAM-D17 remission rate⁵⁸⁾ at Day 15 and 6 weeks later (Day 57). Both endpoints at the both time points tended to favor zuranolone 30 mg over placebo in terms of improvement in depressive symptoms.

58) Proportion of subjects with a HAM-D17 total score of ≤ 7

Table 41. Results of key secondary endpoints (Double-blind phase of Study A3734, FAS)

	Time point	Placebo	Zuranolone 30 mg	Odds ratio [95% CI] ^{a)}
HAM-D17 response rate ^{b)}	Day 15	17.0% (33/194)	22.4% (43/192)	1.40 [0.85, 2.33]
	Day 57	25.5% (47/184)	31.4% (55/175)	1.21 [0.77, 1.93]
HAM-D17 remission rate ^{b)}	Day 15	4.6% (9/194)	7.8% (15/192)	1.74 [0.74, 4.07]
	Day 57	13.6% (25/184)	19.4% (34/175)	1.44 [0.82, 2.54]

a) IPW-GEE with treatment, time point, and treatment-by-time point interaction as fixed effects and baseline HAM-D17 total score, prior drug for depressive episodes (Yes, No), and sex as covariates

b) Proportion (Number of subjects)

Table 42 shows the results of subgroup analyses of the primary endpoint according to patient characteristics during the double-blind phase of Study A3734. No patient characteristics impacting the efficacy of zuranolone were identified based on the results from Study A3734.

Table 42. Change from baseline in HAM-D17 total score at Day 15 by patient characteristics (Double-blind phase of Study A3734, FAS)

Patient characteristics		Treatment group	Baseline	Day 15	Change ^{a) b)}	Treatment difference [95% CI] ^{b)}
Baseline HAM-D17 total score	<25	Placebo	22.8 ± 0.8 (132)	17.3 ± 5.0 (129)	-5.5 ± 0.47	
		Zuranolone 30 mg	23.0 ± 0.8 (136)	16.1 ± 5.7 (128)	-7.0 ± 0.46	-1.49 [-2.78, -0.19]
	≥25	Placebo	26.7 ± 1.6 (67)	18.9 ± 6.7 (65)	-7.6 ± 0.78	
		Zuranolone 30 mg	26.4 ± 1.7 (69)	17.8 ± 5.9 (64)	-8.3 ± 0.78	-0.69 [-2.88, 1.49]
Episode	Single	Placebo	23.9 ± 2.2 (85)	17.8 ± 6.0 (81)	-6.1 ± 0.57	
		Zuranolone 30 mg	23.9 ± 1.5 (84)	17.2 ± 5.1 (80)	-6.7 ± 0.57	-0.66 [-2.25, 0.93]
	Recurrent	Placebo	24.3 ± 2.1 (114)	17.8 ± 5.4 (113)	-6.4 ± 0.57	
		Zuranolone 30 mg	24.3 ± 2.3 (121)	16.3 ± 6.3 (112)	-7.9 ± 0.56	-1.59 [-3.15, -0.02]
Prior treatment with antidepressants	No	Placebo	24.1 ± 2.2 (137)	17.3 ± 5.9 (132)	-6.6 ± 0.52	
		Zuranolone 30 mg	24.0 ± 1.9 (133)	16.7 ± 6.1 (123)	-7.3 ± 0.53	-0.63 [-2.08, 0.82]
	Yes	Placebo	24.3 ± 2.0 (62)	18.9 ± 4.8 (62)	-5.4 ± 0.65	
		Zuranolone 30 mg	24.3 ± 2.0 (72)	16.6 ± 5.4 (69)	-7.7 ± 0.61	-2.38 [-4.14, -0.62]
Prior treatment with anxiolytics	No	Placebo	24.1 ± 2.1 (164)	18.0 ± 5.6 (160)	-6.0 ± 0.45	
		Zuranolone 30 mg	24.2 ± 2.1 (176)	16.9 ± 5.8 (167)	-7.2 ± 0.43	-1.21 [-2.43, 0.01]
	Yes	Placebo	24.4 ± 2.5 (35)	17.1 ± 5.9 (34)	-7.4 ± 1.01	
		Zuranolone 30 mg	23.9 ± 1.3 (29)	15.2 ± 5.8 (25)	-8.7 ± 1.14	-1.32 [-4.38, 1.75]
Prior treatment with hypnotics	No	Placebo	24.0 ± 2.1 (140)	17.5 ± 5.7 (136)	-6.5 ± 0.49	
		Zuranolone 30 mg	24.0 ± 1.8 (136)	16.8 ± 5.9 (130)	-7.3 ± 0.50	-0.82 [-2.19, 0.55]
	Yes	Placebo	24.5 ± 2.2 (59)	18.7 ± 5.3 (58)	-5.7 ± 0.75	
		Zuranolone 30 mg	24.5 ± 2.2 (69)	16.5 ± 5.8 (62)	-7.7 ± 0.70	-2.00 [-4.03, 0.04]

Mean ± SD (Number of evaluable subjects)

a) LS mean ± SE

b) Analysis based on an analysis model similar to the primary analysis model

As to the efficacy of repeated treatment courses with zuranolone, based on the change from baseline in the HAM-D17 total score at Day 15 (the day after the end of dosing) in each 14-day treatment cycle of zuranolone during the open-label extension phase, depressive symptoms tended to improve to a similar extent, regardless of the number of treatment cycles (Table 35), and there was no trend towards a shorter time to next treatment following the completion of the treatment course. Table 43 shows the number of treatment cycles of zuranolone

during the open-label extension phase of Study A3734. The number of subjects who received 6 treatment cycles was highest followed by the number of subjects who received 1 or 2 treatment cycles.

Table 43. Number of treatment cycles of zuranolone during the open-label extension phase of Study A3734

Treatment group during the double-blind phase		Placebo	Zuranolone 30 mg
Number of evaluable subjects		143	128
Number of treatment cycles	1	29 (20.3)	29 (22.7)
	2	20 (14.0)	23 (18.0)
	3	10 (7.0)	13 (10.2)
	4	13 (9.1)	8 (6.3)
	5	15 (10.5)	12 (9.4)
	6	56 (39.2)	43 (33.6)

n (Proportion)

Based on the above, the results from Study A3734 demonstrated the efficacy of zuranolone in the treatment of depressive symptoms.

PMDA's view:

The superiority of zuranolone over placebo was demonstrated for the primary endpoint during the double-blind phase of Study A3734, i.e., the change from baseline in the HAM-D17 total score at Day 15. The secondary endpoints of the HAM-D17 response rate and the HAM-D17 remission rate at Day 15 tended to favor zuranolone 30 mg over placebo. During 6 weeks following the end of study treatment, although the absolute difference between zuranolone and placebo in the change from baseline in the HAM-D17 total score tended to be smaller at Day 22 onwards, the HAM-D17 total score did not tend to worsen over time and tended to favor zuranolone 30 mg over placebo, and a similar trend was observed also for the HAM-D17 response rate and the HAM-D17 remission rate. During the open-label extension phase of Study A3734, also when treatment with zuranolone was repeated, depressive symptoms tended to improve. Based on the above results from Study A3734, the efficacy of zuranolone in the treatment of depressive symptoms was demonstrated.

However, during the open-label extension phase of Study A3734, most subjects received more than 1 treatment course with zuranolone, the majority of whom needed an additional treatment course immediately after 6 weeks off-treatment following the end of dosing. Many subjects received 6 treatment cycles, i.e. the maximum number of treatment cycles. Given these points, the appropriateness of repeated treatment courses with zuranolone will be discussed in Section 7.R.5.2.

7.R.2.2 Efficacy of zuranolone as an add-on to other antidepressants

PMDA asked the applicant to explain the efficacy of zuranolone as an add-on to other antidepressants, based on the results from Study A3736.

The applicant's explanation:

Study A3736 in MDD patients with an inadequate response to ongoing antidepressant monotherapy (the currently approved antidepressants [SSRIs, SNRIs, or vortioxetine hydrobromide]) evaluated the efficacy etc. of zuranolone as an add-on therapy. The point estimate of the adjusted odds ratio of the HAM-D17 response rate at Day 15 for zuranolone vs. placebo (the primary endpoint) was below the pre-specified threshold [see Section 7.3.2], which failed to demonstrate the efficacy of zuranolone as an add-on to other antidepressants.

Although the characteristics of subjects enrolled in Study A3736 and the effects of concomitant antidepressants were examined from various aspects, no clear factors contributing to the above results were identified. Given that the failure rate of placebo-controlled trials for even the currently approved antidepressants is approximately 50% (*Eur Psychiatry*. 2001; 16: 418-23), and that 31.4% of clinical trials for adjunctive therapies for antidepressant partial- and non-responders with MDD were successful (*Mol Psychiatry*. 2020; 25: 1967-74), patients enrolled in Study A3736 may have been resistant to pharmacotherapy, and the study was considered to have a low success rate.

Although Study A3736 failed to demonstrate the efficacy of zuranolone as an add-on to other antidepressants, given that from the standpoint of feasibility, Study A3736 had the primary endpoint different from that of Study A3734 and was not designed to test with sufficient precision the efficacy hypothesis, interpretation of the results of Study A3736 has certain limitations, and it cannot be said that the results rule out the efficacy of zuranolone as an add-on to other antidepressants. In addition, a foreign phase III study to evaluate the efficacy etc. of zuranolone 50 mg in non-Japanese patients with MDD (Study 217-MDD-301B⁴⁰) etc. suggested the efficacy of zuranolone as an add-on to other antidepressants. Zuranolone has a different mechanism of action from other antidepressants based on the monoamine hypothesis, such as SSRIs, SNRIs, or vortioxetine hydrobromide, and a certain level of the add-on effect of zuranolone is expected. There have been no major concerns about the safety of zuranolone as an add-on to other antidepressants [see Section 7.R.3]. Given these points etc., leaving an option of administering zuranolone as an add-on to other antidepressants in clinical practice should have a certain significance.

PMDA's view:

As to the efficacy of zuranolone as an add-on to other antidepressants, the point estimate of the adjusted odds ratio of the HAM-D17 response rate at Day 15 (the primary endpoint of Study A3736) for zuranolone vs. placebo was below the pre-specified threshold, which failed to demonstrate the efficacy of zuranolone as an add-on to other antidepressants. No clear factors contributing to the above results were identified, and the clinical meaningfulness of zuranolone as an add-on to other antidepressants is unknown. On the other hand, given the applicant's explanation that from the standpoint of feasibility, Study A3736 was not designed to test with sufficient precision the efficacy hypothesis, it is also understandable that the results from Study A3736 have certain limitations to evaluating the efficacy of zuranolone as an add-on to other antidepressants. Zuranolone has a different mechanism of action from the currently approved antidepressants based on the monoamine hypothesis, and there have been no major concerns about the safety of zuranolone as an add-on to other antidepressants [see Section 7.R.3]. Given these points etc., there is no need to restrict the use of zuranolone as an add-on to other antidepressants just because of the results from Study A3736.

In Study A3734 with zuranolone monotherapy, the improvements in the HAM-D17 total score were small during 6 weeks off-treatment after the end of dosing, and the absolute difference between zuranolone and placebo in the change from baseline in the HAM-D17 total score tended to be smaller at Day 22 onwards. Based on the above, it is also envisaged that after marketing, in addition to zuranolone, the currently approved antidepressants will be used as treatment intervention for depression. In this case, the option of administering

zuranolone as an add-on to other antidepressants may be considered in clinical practice. Thus, the package insert should advise healthcare professionals to carefully consider the appropriateness of using zuranolone as an add-on to other antidepressants with a full understanding of the results of the efficacy of zuranolone as an add-on to other antidepressants in Study A3736.

7.R.3 Safety

PMDA's conclusion:

Given the following considerations in Sections 7.R.3.1 to 7.R.3.5 based on the submitted study results, attention should be paid to the possible occurrence of CNS adverse events and suicide-related adverse events following administration of zuranolone, and the proper use of zuranolone is important, taking account of the risk of dependence and abuse. Zuranolone should have acceptable safety (including these events) in Japanese patients with depression, as long as zuranolone is used with appropriate precautions.

7.R.3.1 Safety of zuranolone

PMDA asked the applicant to explain the safety of zuranolone based on the occurrence of adverse events in Japanese phase III studies (Studies A3734 and A3736).

The applicant's explanation:

Adverse events reported in Study A3734 with zuranolone monotherapy and Study A3736 with zuranolone administered as an add-on to the currently approved antidepressants such as SSRIs were reviewed. There were no deaths in either study. Among treatment-emergent serious adverse events, those for which a causal relationship to study drug could not be ruled out occurred in 2 subjects during the open-label extension phase of Study A3734 (altered state of consciousness; and liver function test abnormal [1 subject each]), and both events had an outcome of "resolved." The event of altered state of consciousness observed in Study A3734 occurred after the subject took 11 capsules at a time, and this subject did not experience adverse events such as suicide attempt and increased impulsivity. Thus, overdose was potentially associated with the primary disease. The event of liver function test abnormal was potentially associated with alcohol because heavy drinking before the onset of this event was confirmed.

As adverse events characteristic of zuranolone, CNS adverse events such as somnolence and dizziness occurred more frequently in the zuranolone 30 mg group than in the placebo group (Table 36 and Table 39), but no severe events were observed. After repeated dosing of zuranolone, the incidence of adverse events did not tend to increase during the treatment period, and no clinically relevant adverse events such as withdrawal symptoms were reported during the follow-up period or the durability observation period after the end of dosing.

Table 44 shows the incidence of adverse events by total treatment cycles of zuranolone in Study A3734. There was no trend towards increasing incidence of adverse events with increasing number of treatment cycles.

Table 44. Incidence of adverse events by total treatment cycles of zuranolone in Study A3734

	Treatment cycle						
	1	2	3	4	5	6	7
Number of evaluable subjects	348	241	192	160	134	111	43
Adverse events	196 (56.3)	120 (49.8)	77 (40.1)	61 (38.1)	46 (34.3)	34 (30.6)	15 (34.9)
Serious adverse events	1 (0.3)	5 (2.1)	1 (0.5)	0	1 (0.7)	0	0
Adverse events leading to treatment discontinuation	7 (2.0)	3 (1.2)	0	0	0	0	0
Adverse events for which a causal relationship to zuranolone could not be ruled out	110 (31.6)	54 (22.4)	30 (15.6)	21 (13.1)	20 (14.9)	12 (10.8)	5 (11.6)

n (Incidence [%])

Comparison of the incidence of adverse events between Studies A3734 and A3736 (Table 36 and Table 39) showed no major differences in the incidence or nature of adverse events. There was no trend towards occurrence of adverse events specific to zuranolone administered as an add-on to other antidepressants.

PMDA's view:

Given adverse events reported in Studies A3734 and A3736, although adverse events of somnolence, dizziness, etc., tended to occur more frequently, there were no clinically relevant safety concerns. However, taking account of the pharmacological effects of zuranolone and the events reported with benzodiazepines, etc., CNS adverse events, adverse events related to breathing abnormalities, suicide-related adverse events, and adverse events related to dependence and abuse will be assessed in Sections 7.R.3.2 to 7.R.3.5.

7.R.3.2 CNS adverse events

Given that severe sedation was observed in non-clinical studies of zuranolone, etc., PMDA asked the applicant to explain the incidence of CNS adverse events⁵⁹⁾ associated with zuranolone.

The applicant's explanation:

Table 45 shows the incidence of CNS adverse events in Studies A3734 and A3736. In both clinical studies, there was a trend towards a higher incidence in the zuranolone group than in the placebo group, but no serious adverse events were reported. There was no trend towards increasing incidence of adverse events with increasing number of treatment cycles.

Table 45. Incidence of CNS adverse events in Studies A3734 and A3736 (Safety population)

	Study A3734			Study A3736		
	Double-blind phase		Open-label extension phase	Double-blind phase		Open-label extension phase
	Placebo	Zuranolone 30 mg		Placebo	Zuranolone 30 mg	
Number of evaluable subjects	199	205	304	52	55	82
All adverse events	14 (7.0)	49 (23.9)	70 (23.0)	3 (5.8)	14 (25.5)	15 (18.3)
Serious adverse events	0	0	0	0	0	0
Adverse events leading to treatment discontinuation	0	2 (1.0)	3 (1.0)	0	0	2 (2.4)
All adverse events						
Somnolence	12 (6.0)	27 (13.2)	46 (15.1)	2 (3.8)	11 (20.0)	10 (12.2)
Dizziness	3 (1.5)	26 (12.7)	31 (10.2)	1 (1.9)	4 (7.3)	8 (9.8)
Vertigo	0	4 (2.0)	3 (1.0)	0	0	0
Sedation	0	0	1 (0.3)	0	0	1 (1.2)
Dizziness postural	0	0	1 (0.3)	0	0	0
Hypersomnia	0	0	0	1 (1.9)	0	0

n (Incidence [%])

59) MedDRA PTs "sedation," "sedation complication," "somnolence," "hypersomnia," "dizziness," "dizziness postural," "dizziness exertional," "vertigo," "confusional state," "delirium," and "disorientation"

In foreign phase III studies in patients with MDD, in addition to CNS adverse events observed in Studies A3734 and A3736, a few cases of adverse events related to confusional state (confusional state, disorientation, delirium) were reported. As serious adverse events, confusional state occurred in 1 subject (Study 217-MDD-303A⁶⁰⁾), and delirium occurred in 2 subjects (1 subject in Study 217-MDD-301A, 1 subject in Study 217-MDD-303A).

According to the latest foreign post-marketing safety information (as of February 2025),⁶¹⁾ somnolence (115 events), dizziness (75 events), and sedation (31 events) were reported, including serious adverse events of somnolence (3 events), dizziness (3 events), and sedation (2 events).

Based on the above, attention should be paid to the possible occurrence of CNS adverse events of somnolence, dizziness, confusional state, etc. following administration of zuranolone, and the package insert should provide appropriate precautions. Meanwhile, there have been no clinically relevant concerns about the use of zuranolone.

PMDA's conclusion:

Given the results from Japanese and foreign clinical studies etc., attention should be paid to the possible occurrence of adverse events of somnolence, dizziness, confusional state, etc. following administration of zuranolone, and the package insert etc. should appropriately advise about the incidence of those events in clinical studies etc.

7.R.3.3 Adverse events related to breathing abnormalities

Given that excessive sedation accompanied by abnormal breathing was observed in non-clinical studies of zuranolone, etc., PMDA asked the applicant to explain the incidence of adverse events related to breathing abnormalities⁶²⁾ associated with zuranolone.

The applicant's explanation:

In Studies A3734 and A3736, due to the risk of respiratory depression associated with zuranolone, patients with respiratory diseases such as sleep apnoea syndrome were to be excluded.⁶³⁾ No adverse events related to breathing abnormalities were reported in these studies.

Also in foreign phase III studies, no serious adverse events related to breathing abnormalities for which a causal

60) An open-label, uncontrolled study in patients with MDD to evaluate the safety and efficacy of the initial and repeated treatment courses with zuranolone (up to 48 weeks) (Reference data CTD 5.3.5.2-01). Initially, zuranolone 30 mg (a dose reduction to 20 mg was allowed in case of tolerability issues) was to be administered orally once daily after the evening meal for 14 days. However, based on the results of Study 217-MDD-301A, the dose of zuranolone was changed to 50 mg (a dose reduction to 40 mg was allowed in case of tolerability issues) during the study (under Protocol Version 3 and thereafter). Following a 14-day treatment course with zuranolone, if the need for a repeat treatment course was determined, and the minimum 8 weeks had elapsed since the last dose of zuranolone, the subject was considered qualified to receive a repeat treatment course, and a maximum of 5 treatment cycles was allowed.

61) August 4, 2023 to February 3, 2025. An estimated exposure of [REDACTED] patient-years.

62) PTs in the MedDRA SMQ "respiratory failure (broad)"

63) In Studies A3734 and A3736, patients with sleep apnoea syndrome (including suspected sleep apnoea syndrome), interstitial pneumonia, severe bronchial asthma, alveolar hypoventilation syndrome, chronic respiratory failure, pulmonary hypertension, or any other chronic respiratory diseases ineligible for the study in the opinion of a physician were to be excluded.

relationship to zuranolone could not be ruled out were reported. According to the latest foreign post-marketing safety information (as of February 2025),⁶¹⁾ there were no concerns about the respiratory effects of zuranolone.

Based on the above, the risk of respiratory effects of zuranolone should be low. However, given the pharmacological effects of zuranolone, when administering zuranolone to patients with respiratory diseases such as sleep apnoea syndrome, attention should be paid to the possible occurrence of adverse events related to breathing abnormalities.

PMDA's view:

As the results from Japanese and foreign clinical studies etc. raised no concerns about the respiratory effects of zuranolone, the risk of respiratory effects of zuranolone should be low. Given the pharmacological effects of zuranolone, when administering zuranolone to patients with respiratory diseases such as sleep apnoea syndrome, attention should be paid to the possible occurrence of adverse events related to breathing abnormalities.

7.R.3.4 Suicide-related adverse events

Given that patients with depression are at risk of suicide, and that antidepressants have also been reported to increase the risk of suicide, etc., PMDA asked the applicant to explain the incidence of suicide-related adverse events⁶⁴⁾ associated with zuranolone.

The applicant's explanation:

In Studies A3734 and A3736, no suicide-related adverse events were reported. According to assessments using the Columbia Suicide Severity Rating Scale (C-SSRS) during the double-blind phases of Studies A3734 and A3736, the proportions of subjects who did not answer "Yes" to Suicidal Ideation Question 1 (Wish to be Dead) at baseline but answered "Yes" to it at post-baseline assessment (the day after study drug administration onwards) were 2.0% (4 of 199 subjects) in the placebo group and 2.9% (6 of 205 subjects) in the zuranolone group during the double-blind phase of Study A3734 and 1.9% (1 of 52 subjects) in the placebo group and 3.6% (2 of 55 subjects) in the zuranolone group in Study A3736. The proportions of subjects who did not answer "Yes" to Suicidal Ideation Question 2 (Non-Specific Active Suicidal Thoughts) at baseline but answered "Yes" to it at post-baseline assessment were 2.0% (4 of 199 subjects) in the placebo group and 1.5% (3 of 205 subjects) in the zuranolone group in Study A3734 and 0% in the placebo group and 1.8% (1 of 55 subjects) in the zuranolone group in Study A3736. There were no major differences between the placebo and zuranolone groups. C-SSRS assessments during the open-label extension phases of Studies A3734 and A3736 showed no different trend from those during the double-blind phases, and suicidal ideation and behavior did not tend to increase even with increasing number of treatment cycles of zuranolone.

In foreign clinical studies in patients with MDD (Study 217-MDD-201B, Study 217-MDD-301A, Study 217-

64) PTs in the MedDRA SMQ "suicide/self-injury (narrow)"

MDD-301B, Study 217-MDD-305⁶⁵⁾), the incidence of treatment-emergent suicide-related adverse events was 1.7% (15 of 905 subjects). In foreign long-term treatment studies (Study 217-MDD-303A, Study 217-MDD-303B⁶⁶⁾), the incidence of treatment-emergent suicide-related adverse events was 1.3% (18 of 1428 subjects). Given that the incidence of suicide-related adverse events was around 0.4% to 5% in clinical studies of other antidepressants (vortioxetine hydrobromide, venlafaxine hydrochloride, etc.), there was no trend towards a higher incidence of suicide-related adverse events with zuranolone compared with other antidepressants.

According to the latest foreign post-marketing safety information (as of February 2025),⁶¹⁾ 34 suicide-related adverse events were reported, including serious adverse events of suicidal ideation (27 events), completed suicide (1 event), suicide attempt (1 event), and intentional overdose (1 event). None of those events were strongly suspected to be related to zuranolone.

Based on the above, though zuranolone did not tend to increase the risk of suicide, as attention should be paid to the possible occurrence of suicide-related adverse events in the treatment of patients with depression, the package insert should advise about the risk of suicide.

PMDA's conclusion:

Given the results from Japanese and foreign clinical studies etc., there was no trend towards an increased risk of suicide with zuranolone compared with other antidepressants. However, as the possibility that suicide-related adverse events occur in the treatment of patients with depression cannot be ruled out, the package insert should advise about the risk of suicide, and the occurrence of suicide should be watched for also after marketing.

7.R.3.5 Dependence and abuse

Based on the results from non-clinical studies of zuranolone, zuranolone was considered to produce psychological and physical dependence [see Section 5.6.3]. The currently approved benzodiazepines pose the risk of dependence. PMDA asked the applicant to explain the risk of dependence and abuse associated with zuranolone.

The applicant's explanation:

In Studies A3734 and A3736, due to the risk of dependence associated with zuranolone, the duration of use of zuranolone was limited to 14 days, and the treatment could be repeated after an interval of ≥ 6 weeks. Then, no adverse events related to dependence and abuse⁶⁷⁾ were reported in Studies A3734 and A3736. The

65) A placebo-controlled, randomized, double-blind, parallel-group study in patients with MDD aged 18 to 64 years (Reference data CTD 5.3.5.1-07). The key inclusion criteria were patients with depressive symptoms persisting for ≥ 4 weeks and a HAM-D17 total score of ≥ 24 . Placebo or zuranolone was to be co-initiated with SSRI or SNRI: Placebo or zuranolone 50 mg (a dose reduction to 40 mg was allowed in case of tolerability issues) was to be administered orally once daily after the evening meal for 14 days, and SSRI or SNRI was to be administered orally for 42 days.

66) An open-label, uncontrolled study in patients with MDD to evaluate the safety and efficacy of the initial and repeated treatment courses with zuranolone (up to 48 weeks) (Reference data CTD 5.3.5.2-02). Zuranolone 50 mg (a dose reduction to 40 mg was allowed in case of tolerability issues) was to be administered orally once daily after the evening meal for 14 days. Following a 14-day treatment course with zuranolone, if the need for a repeat treatment course was determined, and the minimum 8 weeks had elapsed since the last dose of zuranolone, the subject was considered qualified to receive a repeat treatment course, and a maximum of 4 treatment cycles was allowed.

67) PTs in the MedDRA SMQ "drug abuse, dependence and withdrawal (broad)"

Dependence Assessment Committee⁶⁸⁾ assessed the risk of dependence and abuse, including the dependence questionnaire,⁶⁹⁾ and found no cases of evident drug dependence or abuse.

Foreign clinical studies and the latest foreign post-marketing safety information (as of February 2025)⁶¹⁾ raised no concerns about dependence and abuse of zuranolone.

Based on the above, though Japanese and foreign clinical studies etc. raised no clear concerns about dependence and abuse of zuranolone, as zuranolone was considered to produce psychological and physical dependence based on non-clinical studies, and a human abuse potential study of zuranolone (Study 217-CLP-110) suggested that zuranolone is associated with abuse potential similar to or less than alprazolam [see Section 6.2.5.2], etc., zuranolone is considered to have a potential risk of dependence. Thus, the package insert should include a precautionary statement about the risk of dependence and advise against repeating treatment with zuranolone indiscriminately.

PMDA's view:

Given the results from non-clinical studies and a human abuse potential study, zuranolone is considered to have the risk of dependence. Meanwhile, since Japanese and foreign clinical studies etc. raised no clear concerns about dependence and abuse of zuranolone in humans, the risk of dependence and abuse should be low with the proposed dosing regimen of zuranolone (zuranolone 30 mg taken orally once daily after the evening meal for 14 days and re-treatment after a minimum interval of 6 weeks) at present. However, given the risk of dependence associated with zuranolone, it is important to use zuranolone in eligible patients, adhere to the dosage and administration, and avoid repeating treatment with zuranolone indiscriminately. Thus, the package insert etc. should include the relevant precautionary statements, and an effort should be made to ensure the proper use of zuranolone. It is also necessary to collect post-marketing information on dependence and abuse.

7.R.4 Target population, clinical positioning, and indications

PMDA asked the applicant to explain the target population and clinical positioning of zuranolone, taking account of the results from Study A3734 etc. and the treatment paradigm for depression in Japan.

The applicant's explanation:

For the treatment of depression in Japan, active pharmacotherapy is not recommended for patients with mild depressive symptoms, and non-pharmacotherapy such as psychotherapy is considered. The use of pharmacotherapy is considered for patients with moderate or severe depression. In Japan, antidepressants based on the monoamine hypothesis such as SSRIs, SNRIs, and vortioxetine hydrobromide have been approved. When these antidepressants are used as pharmacotherapy, adequate continuation and maintenance therapy should be conducted to prevent relapse/recurrence after the remission of depressive symptoms achieved in the

68) The Dependence Assessment Committee constituted of 3 external psychiatrists was to assess cases of adverse events of dependence and abuse and subjects who chose "quite a lot" or "moderately" for at least 1 item of the dependence questionnaire, reviewing individual data, in accordance with the operating procedures.

69) Using a 10-item questionnaire to assess drug dependence and abuse (D-2-A) and a 6-item questionnaire to assess withdrawal symptoms (D-2-B), subjects were interviewed, and each question was rated on a 4-point scale: "quite a lot, moderately, slightly, and not at all."

acute phase of treatment, and pharmacotherapy should not be terminated too quickly (Japanese Guideline for Treatment of Depression). In the acute phase treatment with antidepressants such as SSRIs, around 8 weeks of treatment are required to assess patient response. Thus, patients may drop out of therapy before actually feeling its effect, and a slow onset of action is an issue for pharmacotherapy (*Japanese Journal of Clinical Neuropsychopharmacology*. 2009; 12: 1703-8).

Zuranolone is expected to exert antidepressant effects by enhancing GABA_A receptor function through a different mechanism from other antidepressants based on the monoamine hypothesis such as SSRIs [see Section 3.R.1]. Due to the risk of dependence, zuranolone was developed as an antidepressant intended for acute phase treatment, and Japanese clinical studies were conducted. A Japanese phase III study (1) (Study A3734) demonstrated the superiority of zuranolone over placebo in the primary endpoint of the change from baseline in the HAM-D17 total score at Day 15, etc., showing that zuranolone has efficacy [see Section 7.R.2] and acceptable safety [see Section 7.R.3]. Thus, zuranolone will become a new treatment option that is expected to provide a more rapid onset of action than the currently approved antidepressants in the acute phase treatment of depression.

PMDA asked the applicant to explain the appropriateness of the proposed indications, taking account of the target population and clinical positioning of zuranolone.

The applicant's explanation:

While Study A3734 etc. demonstrated the efficacy of zuranolone, its long-term continuous use should be avoided due to the risk of dependence. Thus, the target population for zuranolone should be patients diagnosed with a first episode of depression with symptoms or patients with relapse/recurrence of depressive symptoms requiring treatment intervention, and zuranolone should not be used to prevent relapse/recurrence.

Then, as to the indications for zuranolone, the target population is patients with depression requiring acute phase treatment, which is similar to those of the currently approved antidepressants such as SSRIs, except that zuranolone should not be used to prevent relapse/recurrence. Although specifying in the INDICATIONS section that zuranolone should be used for acute phase treatment only was also considered, as the definition of acute phase is not necessarily clear, some physicians may think that zuranolone is indicated for severe or urgent patients, and the target population may not be understood appropriately. Thus, the indications for zuranolone should be "depression and depressive state," and the package insert etc. should advise against the use of zuranolone for the prevention of relapse/recurrence.

PMDA's view:

Since Study A3734 etc. demonstrated the efficacy of zuranolone in the treatment of depressive symptoms [see Section 7.R.2] and its acceptable safety [see Section 7.R.3], zuranolone will become an acute phase treatment option for patients with depression. Given the risk of dependence associated with zuranolone, its long-term continuous use should be avoided, and zuranolone use for acute phase treatment only and not for treatment in continuation and maintenance phases to prevent relapse/recurrence, is justified. It is important to correctly and

appropriately provide the following information on the target population to healthcare professionals in clinical practice: Unlike other antidepressants based on the monoamine hypothesis such as SSRIs, zuranolone should not be used to prevent relapse/recurrence.

Then, given the target population for zuranolone and the applicant's explanation, the proposed indications of "depression and depressive state" are appropriate, and the package insert etc. should advise against the use of zuranolone for the prevention of relapse/recurrence.

The above conclusion by PMDA will be finalized, taking account of comments from the Expert Discussion.

7.R.5 Dosage and administration

7.R.5.1 Dosage and administration for zuranolone

The applicant's explanation about the appropriateness of the proposed dosing regimen of zuranolone:

The results from a Japanese phase II study (Study A3731) suggested the efficacy of zuranolone at both doses of 20 mg and 30 mg, and there were no major differences in safety between zuranolone 20 mg and 30 mg. Meanwhile, based on the results of other efficacy endpoints as well as the primary endpoint, higher efficacy was expected with zuranolone 30 mg than with 20 mg, etc. Thus, a dose of 30 mg was selected for Japanese phase III studies (Studies A3734 and A3736), and zuranolone 30 mg was to be administered orally once daily after the evening meal for 14 days. Treatment with zuranolone could be started in the open-label extension phase after ≥ 6 weeks off-treatment following the end of dosing. As to the timing of dosing, since food affected the pharmacokinetics of zuranolone [see Section 6.R.1], and somnolence, dizziness, etc. occurred as adverse events of zuranolone [see Section 7.R.3.2], zuranolone was to be administered after the evening meal in Japanese clinical studies etc. The study results demonstrated the efficacy of a 14-day administration of zuranolone in the treatment of depressive symptoms and showed that a certain level of efficacy is expected with no reduced efficacy even after repeated treatment courses with zuranolone [see Section 7.R.2]. Zuranolone had acceptable safety [see Section 7.R.3].

Based on the above, the appropriate dosing regimen is as follows: Zuranolone 30 mg should be administered orally once daily after the evening meal for 14 days, and the treatment may be repeated after a minimum interval of 6 weeks.

7.R.5.2 Treatment after completion of the 14-day treatment course with zuranolone

Given that zuranolone should be used for acute phase treatment only due to the risk of dependence, and that 14 days of treatment with zuranolone is followed by ≥ 6 weeks off-treatment, PMDA asked the applicant to explain treatment of patients with depression following the completion of the 14-day treatment course with zuranolone.

The applicant's explanation:

As to treatment following the completion of the 14-day treatment course with zuranolone, it is desirable to check the patient's condition at regular visits every 1 to 2 weeks also after the end of dosing. Taking account of the clinical course, psychosocial factors, etc., of each patient, non-pharmacotherapy such as psychotherapy or switching to other antidepressants will be selected via Shared Decision Making between healthcare professionals and patients. It is envisaged that zuranolone will be switched to other drugs if depressive symptoms do not improve adequately even after the end of dosing or if depressive symptoms improve following administration of zuranolone, and non-pharmacotherapy such as psychotherapy is performed, but the symptoms worsen during an off-treatment period, i.e., within 6 weeks after the end of dosing. On the other hand, if depressive symptoms improve and are milder or remit following 14 days of treatment with zuranolone and do not relapse or recur during 6 weeks off-treatment following the end of dosing, patient monitoring and treatment with non-pharmacotherapy etc. will be continued, and if pharmacotherapy is required due to relapse/recurrence, treatment with zuranolone or treatment with other drugs will be selected. If treatment with zuranolone is selected, the above-mentioned decision making will be repeated.

PMDA asked the applicant to explain the appropriateness of repeated treatment courses with zuranolone in patients with depression.

The applicant's explanation:

During the open-label extension phase of Study A3734, depressive symptoms tended to improve to a similar extent, regardless of the number of treatment cycles, and there was no trend towards a shorter time to next treatment following the completion of the treatment course [see Section 7.R.2]. A certain number of patients remitted (a HAM-D17 total score ≤ 7) following repeated treatment courses with zuranolone. There were no clinically relevant safety concerns about repeated treatment courses with zuranolone [see Section 7.R.3]. Given these points etc., there should be no problems with repeated treatment courses with zuranolone, as long as patients remain off-treatment for 6 weeks after the end of dosing. However, in patients who are likely to relapse following treatment with zuranolone in the acute phase only, e.g., patients who tend to relapse based on their history of depression or prior treatment etc., treatment with zuranolone should not be repeated and should be switched to other therapies.

PMDA's view based on the applicant's explanation in Sections 7.R.5.1 and 7.R.5.2:

Dosage and administration for zuranolone

Given the results from Study A3734 etc., which were conducted on the premise that zuranolone is used for acute phase treatment only due to the risk of dependence, zuranolone 30 mg should be administered orally once daily after the evening meal for 14 days, and if relapse/recurrence occurs, the treatment may be repeated after a minimum interval of 6 weeks.

Treatment following the completion of the 14-day treatment course with zuranolone

As explained by the applicant, it is important to regularly monitor patients' condition and clinical course even after completion of a treatment course with zuranolone, to select either non-pharmacotherapy such as

psychotherapy or pharmacotherapy involving a switch to another antidepressant as appropriate for each patient, and to continue treatment. However, due to the risk of dependence, adherence to the dosage and administration (14 days of treatment with zuranolone followed by ≥ 6 weeks off-treatment) is extremely important, and treatment with zuranolone should not be repeated indiscriminately. Moreover, many subjects received an additional treatment course immediately after 6 weeks off-treatment during the open-label extension phase of Study A3734, and there is a possibility that such patients did not achieve complete remission of depressive symptoms despite repeated treatment courses with zuranolone. Thus, the package insert should advise that other therapies should be considered for patients who continue to experience repeated relapses/recurrences despite treatment with zuranolone.

The above conclusion by PMDA will be finalized, taking account of comments from the Expert Discussion.

7.R.6 Post-marketing investigations

Based on the considerations on the safety of zuranolone [see Section 7.R.3], the applicant considers that attention should be paid to the possible occurrence of somnolence, dizziness, etc. following administration of zuranolone and plans a general use-results survey primarily to investigate the incidences of these events in clinical practice.

PMDA considers that there is no major problem with the applicant's measures, but will finalize the details of the risk management plan including the post-marketing surveillance plan, taking account of comments from the Expert Discussion.

8. Results of Compliance Assessment Concerning the New Drug Application Data and Conclusion Reached by PMDA

8.1 PMDA's conclusion concerning the results of document-based GLP/GCP inspections and data integrity assessment

The new drug application data were subjected to a document-based inspection and a data integrity assessment in accordance with the provisions of the Act on Securing Quality, Efficacy and Safety of Products Including Pharmaceuticals and Medical Devices. On the basis of the inspection and assessment, PMDA concluded that there were no obstacles to conducting its review based on the application documents submitted.

8.2 PMDA's conclusion concerning the results of the on-site GCP inspection

The new drug application data (CTD 5.3.5.1-02) were subjected to an on-site GCP inspection, in accordance with the provisions of the Act on Securing Quality, Efficacy and Safety of Products Including Pharmaceuticals and Medical Devices. On the basis of the inspection, PMDA concluded that there were no obstacles to conducting its review based on the application documents submitted.

9. Overall Evaluation during Preparation of the Review Report (1)

On the basis of the data submitted, PMDA has concluded that zuranolone has efficacy in the treatment of depression and depressive state, and that zuranolone has acceptable safety in view of its benefits. The drug

product is classified as a powerful drug, and its drug substance is classified as a poisonous drug. Zuranolone is clinically meaningful because it offers a treatment option for patients with depression or depressive state.

PMDA has concluded that zuranolone may be approved if zuranolone is not considered to have any particular problems based on comments from the Expert Discussion.

Review Report (2)

October 8, 2025

Product Submitted for Approval

Brand Name	Zurzuvae Capsules 30 mg
Non-proprietary Name	Zuranolone
Applicant	Shionogi & Co., Ltd.
Date of Application	September 27, 2024

List of Abbreviations

See Appendix.

1. Content of the Review

Comments made during the Expert Discussion and the subsequent review conducted by the Pharmaceuticals and Medical Devices Agency (PMDA) are summarized below. The expert advisors present during the Expert Discussion were nominated based on their declarations, etc. concerning the product submitted for marketing approval, in accordance with the provisions of the Rules for Convening Expert Discussions, etc. by Pharmaceuticals and Medical Devices Agency (PMDA Administrative Rule No. 8/2008 dated December 25, 2008).

At the Expert Discussion, the expert advisors supported PMDA's conclusion concerning the safety of zuranolone presented in the Review Report (1).

1.1 Efficacy, target population, clinical positioning, indications, and dosage and administration

1.1.1 Efficacy, target population, clinical positioning, and indications

The expert advisors made the following comments on the efficacy, target population, clinical positioning, and indications of zuranolone and supported PMDA's conclusions presented in Sections "7.R.2 Efficacy" and "7.R.4 Target population, clinical positioning, and indications" in the Review Report (1).

- Study A3734 demonstrated the superiority of zuranolone over placebo in the primary endpoint during the double-blind phase of the change from baseline in the HAM-D17 total score at Day 15 (Table 33), and the secondary endpoints of the HAM-D17 response rate and the HAM-D17 remission rate at Day 15 tended to favor zuranolone 30 mg over placebo (Table 41). Given these findings, PMDA's conclusion that the efficacy of zuranolone in the treatment of depressive symptoms was demonstrated is appropriate. Since the zuranolone 30 mg group tended to have a greater decrease in the HAM-D17 total score compared to placebo at Days 3 and 8 during the double-blind phase of Study A3734 (Table 34), zuranolone is expected to provide rapid onset of action, unlike the currently approved antidepressants such as SSRIs.
- According to Study A3736, which was conducted to evaluate the efficacy of zuranolone as an add-on to other antidepressants, the point estimate of the adjusted odds ratio of the primary endpoint of the HAM-

D17 response rate at Day 15 for zuranolone vs. placebo was below the pre-specified threshold [see Section 7.3.2]. Moreover, with respect to the change from baseline in the HAM-D17 total score over time during the double-blind phase of Study A3736 (Table 38), there was a trend towards inferior efficacy in the zuranolone group compared to the placebo group at all time points. Based on the above, even considering that Study A3736 was not designed to test the efficacy hypothesis with sufficient precision, the study results appear to rule out at least a substantial effect of zuranolone as an add-on to other antidepressants. PMDA's conclusion that, after marketing, there is no need to restrict the use of zuranolone as an add-on to other antidepressants, that information on the results from Study A3736 should be provided through the package insert and materials for healthcare professionals, and that healthcare professionals should be advised to carefully consider the appropriateness of such use with a full understanding of the results of the efficacy of zuranolone as an add-on to other antidepressants in Study A3736, is appropriate.

- The following conclusion by PMDA is appropriate:

Due to the risk of dependence, long-term continuous use of zuranolone should be avoided, and zuranolone should not be used to prevent relapse/recurrence and should be used for acute phase treatment only. Given these points, the indications should be "depression and depressive state," and the package insert should advise against the use of zuranolone for the prevention of relapse/recurrence.

1.1.2 Dosage and administration

The expert advisors made the following comments on the dosing regimen of zuranolone and supported PMDA's conclusion presented in Section "7.R.5 Dosage and administration" in the Review Report (1).

- Dosage and administration

PMDA's conclusion that, given the results from Study A3734, zuranolone 30 mg should be administered orally once daily after the evening meal for 14 days and that treatment may be repeated after a minimum interval of 6 weeks is appropriate.

- Treatment following the completion of the 14-day treatment course with zuranolone

PMDA's conclusion that it is important to regularly monitor patients' condition and clinical course even after completion of a treatment course with zuranolone, to select either non-pharmacotherapy such as psychotherapy or pharmacotherapy involving a switch to another antidepressant as appropriate for each patient, and to continue treatment, is appropriate. Furthermore, given that most subjects received multiple treatment courses with zuranolone during the open-label extension phase of Study A3734 and that the largest proportion of subjects received 6 treatment courses (Table 43), while zuranolone provides a rapid onset of action, treatment with zuranolone may not necessarily be sufficient to achieve complete remission without relapse/recurrence. Therefore, PMDA's conclusion that healthcare professionals should be advised that, due to the risk of dependence, adherence to the dosage and administration is extremely important, that treatment with zuranolone should not be repeated indiscriminately, and that other therapies should be considered for patients who continue to experience repeated relapses/recurrences despite treatment with zuranolone, is appropriate.

In view of the comments from the Expert Discussion in Sections 1.1.1 and 1.1.2, PMDA concluded as follows: The proposed indications for zuranolone are appropriate, and the following statements should be included in

the PRECAUTIONS CONCERNING INDICATIONS section of the package insert. The dosage and administration statement should be modified as follows, and the following statements should be included in the PRECAUTIONS CONCERNING DOSAGE AND ADMINISTRATION section. Then, information should be provided appropriately to healthcare professionals and patients, using information materials so that the target population, clinical positioning, and treatment approach are clear for the proper use of zuranolone.

PMDA instructed the applicant about the above points, and the applicant responded accordingly.

Indications (No change from the proposed text.)

Depression and depressive state

Precautions Concerning Indications (excerpt)

Administer zuranolone to patients with depressive symptoms. Do not administer zuranolone for the prevention of relapse/recurrence in patients whose depressive symptoms have remitted or resolved.

Dosage and Administration (The underlined words are added to the proposed text, and the strikethrough words are deleted from the proposed text.)

The usual adult dosage is 30 mg zuranolone taken orally once daily after the evening meal for 14 days. The treatment may be repeated after a minimum interval of at least 6 weeks ~~after~~ from the end of dosing.

Precautions Concerning Dosage and Administration

- Due to the risk of drug dependence, adhere to the dosage and administration and carefully consider the therapeutic need for repeated treatment with zuranolone.
- If relapse/recurrence occurs after remission or resolution of depressive symptoms following 14 days of treatment with zuranolone, the treatment may be repeated only after a minimum interval of 6 weeks from the end of dosing. Other therapies should be considered for patients who relapse/recur despite repeated treatment courses with zuranolone, and treatment with zuranolone should not be repeated indiscriminately.
- If depressive symptoms do not improve following 14 days of treatment with zuranolone or if depressive symptoms worsen within 6 weeks after the end of dosing and require pharmacotherapy, other therapies should be considered, e.g., switching to other antidepressants, instead of re-treatment with zuranolone.
- Even if depressive symptoms remit or resolve following 14 days of treatment with zuranolone, the patient's condition should be monitored regularly, and appropriate treatment should be performed according to the patient's condition, e.g., non-pharmacotherapy such as psychotherapy.
- If zuranolone is used in patients on other antidepressants, zuranolone monotherapy should be considered. Whether to use zuranolone as an add-on to other antidepressants should be decided carefully, with a full understanding of the information presented in Section "17. CLINICAL STUDIES" and of the efficacy and safety of zuranolone.

1.2 Risk management plan (draft)

At the Expert Discussion, the expert advisors supported PMDA's conclusion presented in Section "7.R.6 Post-marketing investigations" in the Review Report (1). PMDA has concluded that the risk management plan (draft) for zuranolone should include the safety specification presented in Table 46, and that the applicant should conduct additional pharmacovigilance activities and risk minimization activities presented in Table 47 and a general use-results survey presented in Table 48.

Table 46. Safety and efficacy specifications in the risk management plan (draft)

Safety specification		
Important identified risks	Important potential risks	Important missing information
· Somnolence/dizziness, etc. · Confusional state	· Excessive sedation · Dependence/abuse · Suicidal ideation/suicidal behavior · Embryo-fetal toxicity	· None
Efficacy specification		
· None		

Table 47. Summary of additional pharmacovigilance activities and additional risk minimization activities included under the risk management plan (draft)

Additional pharmacovigilance activities	Additional risk minimization activities
· Early post-marketing phase vigilance · General use-results survey	· Dissemination of information obtained through early post-marketing phase vigilance · Preparation and distribution of information materials for healthcare professionals · Preparation and distribution of information materials for patients

Table 48. Outline of general use-results survey plan (draft)

Objective	To investigate the safety and efficacy of zuranolone in clinical practice.
Survey method	Central registry system
Population	Patients with depression/depressive state
Observation period	52 weeks
Planned sample size	1500 patients
Main survey items	· Patient characteristics (age, sex, the number of depressive episodes, complication, etc.) · Use of zuranolone (including the number of treatment cycles of zuranolone) · Use of other therapeutic drugs for depression/depressive state · Non-pharmacotherapy for depression/depressive state used during the observation period · Adverse events · Depressive symptoms (HAM-D17, PHQ-9, etc.)

2. Overall Evaluation

As a result of the above review, PMDA has concluded that the product may be approved after modifying the proposed indications and dosage and administration as shown below, with the following condition. Since the product is a drug with a new active ingredient, the re-examination period is 8 years.

Indications

Depression and depressive state

Dosage and Administration

The usual adult dosage is 30 mg zuranolone taken orally once daily after the evening meal for 14 days. The treatment may be repeated after a minimum interval of 6 weeks from the end of dosing.

Approval Condition

The applicant is required to develop and appropriately implement a risk management plan.

List of Abbreviations

ALT	Alanine aminotransferase
APTT	Activated partial thromboplastin time
AST	Aspartate aminotransferase
AUC	Area under concentration-time curve
BCRP	Breast cancer resistance protein
BSEP	Bile salt export pump
CDP	Chlordiazepoxide
CHO	Chinese hamster ovary
CI	Confidence interval
C _{max}	Maximum concentration
COVID-19	Coronavirus disease 2019
CPP	Critical process parameter
CQA	Critical quality attribute
C-SSRS	Columbia Suicide Severity Rating Scale
CTD	Common technical document
C _{trough}	Trough concentration
DI	Deionized
DMSO	Dimethyl sulfoxide
DSM-5	Diagnostic and Statistical Manual of Mental Disorders Fifth Edition
EC ₅₀	50% effective concentration
ED ₅₀	50% effective dose
EEG	Electroencephalography
eGFR	Estimated glomerular filtration rate
E _{max}	Maximum effect
FAS	Full analysis set
FOB	Functional observational battery
GABA	γ -aminobutyric acid
GC	Gas chromatography
GCP	Good Clinical Practice
HAM-D17	Hamilton rating scale for depression
HEK	Human embryonic kidney
hERG	human ether-à-go-go related gene
HP β CD	Hydroxypropyl- β -cyclodextrin
HPLC	High performance liquid chromatography
IC ₅₀	50% inhibitory concentration
ICH Q1E guideline	"Guideline on Evaluation of Stability Data" (PMSB/ELD Notification No. 0603004 dated June 3, 2003)
IPSC	Inhibitory postsynaptic current
IPW-GEE	Inverse probability-weighted generalized estimating equation
IR	Infrared absorption spectroscopy
Japanese Guideline for Treatment of Depression	Treatment Guideline II by the Japanese Society of Mood Disorders. Depression (DSM-5)/Major Depressive Disorder 2016, revised in 2024. Drafting Committee for the Treatment Guideline for Mood Disorders, the Japanese Society of Mood Disorders.
K _i	Inhibitory constant
LC-MS/MS	Liquid chromatography-tandem mass spectrometry
Ltk	Mouse fibroblast cell line
MATE	Multidrug and toxin extrusion protein
MDCK	Madin-Darby canine kidney

MDD	Major depressive disorder
MedDRA	Medical dictionary for regulatory activities
mFAS	Modified full analysis set
MMRM	Mixed-effects model repeated measures
mRNA	Messenger ribonucleic acid
NMR	Nuclear magnetic resonance spectroscopy
NZW	New Zealand White
OAT	Organic anion transporter
OATP	Organic anion transporting polypeptide
OCT	Organic cation transporter
P450	Cytochrome P450
PAM	Positive allosteric modulator
PAPS	3'-phosphoadenosine-5'-phosphosulfate
PAR	Proven acceptable range
PEG	Polyethylene glycol
P-gp	P-glycoprotein
PHQ-9	Patient Health Questionnaire-9
PMDA	Pharmaceuticals and Medical Devices Agency
PPK	Population pharmacokinetics
PT	Preferred term
PTP	Press through packaging
PVP-VA	Polyvinylpyrrolidone-vinyl acetate
QTc	Corrected QT
QTcF	QT interval corrected for heart rate using Fridericia's method
RH	Relative humidity
SD	Sprague Dawley
SDLP	Standard Deviation of Lateral position
SMQ	Standardized MedDRA query
SNRI	Serotonin-noradrenaline reuptake inhibitor
SSRI	Selective serotonin reuptake inhibitor
$t_{1/2}$	Elimination half-life
Tg	Transgenic
The product	Zurzuva Capsules 30 mg
t_{max}	Time to reach maximum concentration
TRPV1	Transient receptor potential cation channel subfamily V member 1
UV-VIS	Ultraviolet-visible spectroscopy
VAS	Visual analogue scale
Zuranolone	zuranolone