

Report on the Deliberation Results

March 15, 2013

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

[Brand name]	Nourias Tablets 20 mg
[Non-proprietary name]	Istradefylline (JAN*)
[Applicant]	Kyowa Hakko Kirin Co., Ltd.
[Date of application]	March 30, 2012

[Results of deliberation]

In the meeting held on March 8, 2013, the First Committee on New Drugs concluded that the product may be approved and that this result should be reported to the Pharmaceutical Affairs Department of the Pharmaceutical Affairs and Food Sanitation Council.

The product is not classified as a biological product or a specified biological product, the re-examination period is 8 years, and neither the drug substance nor the drug product is classified as a poisonous drug or a powerful drug.

**Japanese Accepted Name (modified INN)*

This English version of the Japanese review report is intended to be a reference material to provide convenience for users. In the event of inconsistency between the Japanese original and this English translation, the former shall prevail. The PMDA will not be responsible for any consequence resulting from the use of this English version.

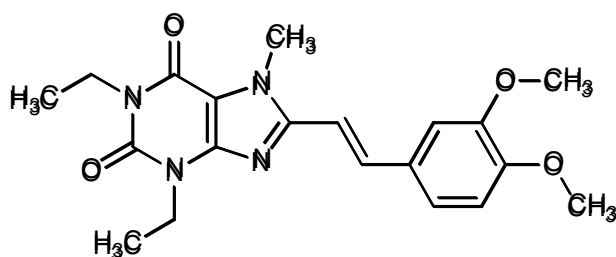
Review Report

February 22, 2013

Pharmaceuticals and Medical Devices Agency

The results of a regulatory review conducted by the Pharmaceuticals and Medical Devices Agency on the following pharmaceutical product submitted for registration are as follows.

[Brand name] Nourias Tablets 20 mg
[Non-proprietary name] Istradefylline
[Name of applicant] Kyowa Hakko Kirin Co., Ltd.
[Date of application] March 30, 2012
[Dosage form/Strength] Film-coated tablets containing 20 mg as Istradefylline per tablet
[Application classification] Prescription drug (1) Drug with a new active ingredient
[Chemical structure]



Molecular formula: C₂₀H₂₄N₄O₄

Molecular weight: 384.43

Chemical name: (E)-8-(3,4-Dimethoxystyryl)-1,3-diethyl-7-methyl-3,7-dihydro-1H-purine-2,6-dione

[Items warranting special mention] None
[Reviewing office] Office of New Drug II

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Review Results

February 22, 2013

[Brand name]	Nourias Tablets 20 mg
[Non-proprietary name]	Istradefylline
[Name of applicant]	Kyowa Hakko Kirin Co., Ltd.
[Date of application]	March 30, 2012

[Results of review]

Based on the submitted data, the efficacy of the product in improving the “wearing-off” phenomenon in patients with Parkinson’s disease on levodopa-containing preparations has been demonstrated and its safety is considered acceptable in view of its observed benefits. It is necessary to collect post-marketing information on the occurrence of psychiatric symptoms and dyskinesias, safety and efficacy in patients treated with 40 mg of istradefylline, safety in smokers, patients with hepatic impairment, patients with ischemic heart disease, and patients with respiratory disorders, and potential risks such as psychological dependence and lung toxicity.

As a result of its regulatory review, the Pharmaceuticals and Medical Devices Agency has concluded that the product may be approved for the following indication and dosage and administration.

[Indication]

Improvement of the “wearing-off” phenomenon in patients with Parkinson’s disease on levodopa-containing preparations

[Dosage and administration]

The product should be used in combination with levodopa-containing preparations. The usual adult dosage is 20 mg of Istradefylline orally administered once daily. According to symptoms, 40 mg of Istradefylline may be orally administered once daily.

Review Report (1)

January 8, 2013

I. Product Submitted for Registration

[Brand name] Nourias Tablets 20 mg
[Non-proprietary name] Istradefylline
[Name of applicant] Kyowa Hakko Kirin Co., Ltd.
[Date of application] March 30, 2012
[Dosage form/Strength] Film-coated tablets containing 20 mg as Istradefylline per tablet
[Proposed indication]

Patients with Parkinson's disease on levodopa-containing preparations with motor complications

[Proposed dosage and administration]

The product should be used in combination with levodopa-containing preparations. The usual adult dosage is 20 mg of Istradefylline orally administered once daily. The dose should be increased as appropriate according to symptoms and 40 mg of Istradefylline may be orally administered once daily.

II. Summary of the Submitted Data and Outline of the Review by the Pharmaceuticals and Medical Devices Agency

The data submitted in the application and the outline of a review by the Pharmaceuticals and Medical Devices Agency (PMDA) are as shown below.

1. Origin or history of discovery and usage conditions in foreign countries etc.

Parkinson's disease (PD) is a progressive neurodegenerative disorder characterized by motor deficits including akinesia, rigidity, bradykinesia, and postural reflex impairment, resulting from the degeneration and loss of dopaminergic neurons projecting from the midbrain substantia nigra pars compacta to the striatum and a subsequent reduction in striatal dopamine content.

Due to dopamine D₂ receptor-mediated inhibitory control over the GABAergic (γ-aminobutyric acid) output of the indirect striato-pallidal pathway, in Parkinson's disease, there is a reduction in dopamine content, which is considered to lead to a loss of dopaminergic input and an enhanced GABAergic output. A selective adenosine A_{2A} receptor antagonist, istradefylline, antagonizes adenosine A_{2A} receptors expressed in GABAergic medium spiny neurons of the indirect striato-pallidal pathway and is expected to improve the symptoms of Parkinson's disease by reducing the overactive GABAergic output of the indirect pathway, i.e. through a mechanism different from conventional anti-PD medications that act on dopamine receptors or dopamine-metabolizing enzymes. Although istradefylline was developed as an anti-PD medication by Kyowa Hakko Kirin Co., Ltd. and a new drug application was filed in 2007 in the US,

[REDACTED]
[REDACTED]
[REDACTED]. As of March 2012, istradefylline is not approved or marketed

in any country of the world.

In Japan, Kyowa Hakko Kogyo Co., Ltd. (a predecessor of Kyowa Hakko Kirin Co., Ltd.) undertook the development of istradefylline tablets in 1996 and based on the data from Japanese clinical studies etc., a marketing application for istradefylline tablets indicated for “patients with Parkinson’s disease on levodopa-containing preparations with motor complications” has now been submitted.

2. Data relating to quality

2.A Summary of the submitted data

2.A.(1) Drug substance

2.A.(1).1 Characterization

The drug substance is a light yellow-green needle crystal and has been characterized by description, powder X-ray diffraction, melting point, thermal analysis, solubility, pH, hygroscopicity, acid dissociation constant ([REDACTED]), and partition coefficient.

The chemical structure of the drug substance has been confirmed by ultraviolet-visible spectroscopy (UV), infrared spectrophotometry (IR), nuclear magnetic resonance spectrometry (¹H-NMR, ¹³C-NMR), mass spectrometry (MS), elementary analysis, and single-crystal x-ray crystallography.

2.A.(1).2 Manufacturing process

The drug substance is synthesized using [REDACTED]
[REDACTED] as starting materials.

[REDACTED] step and [REDACTED] step have been defined as critical steps.
[REDACTED].

2.A.(1).3 Control of drug substance

The proposed specification for the drug substance include content, description (visual), identification (UV, IR), purity (heavy metals [method 2 of the heavy metals limit test], related substances [liquid chromatography (HPLC)]), residue on ignition (residue on ignition test), [REDACTED], and assay (HPLC).

2.A.(1).4 Stability of drug substance

The stability studies conducted on the drug substance are as shown in Table 1. Photostability data showed that the drug substance is photolabile.

Table 1. Stability studies on drug substance

Study	Primary batches	Temperature	Humidity	Storage package	Storage period
Long-term	3 production batches	25°C	60%RH	double polyethylene bags + fiber drums (protected from light)	60 months
Accelerated		40°C	75%RH		6 months

Based on “Guideline on Evaluation of Stability Data” (PMSB/ELD Notification No. 0603004 dated June 3, 2003, “ICH Q1E Guideline”), a re-test period of 60 months has been proposed for the drug substance when stored in double polyethylene bags within fiber drums, protected from light, at room temperature.

2.A.(2) Drug product

2.A.(2).1 Description and composition of the drug product

The drug product is presented as yellow-brown film-coated tablets containing 20 mg of istradefylline per tablet. It contains lactose hydrate, microcrystalline cellulose, crospovidone, polyvinyl alcohol (partially hydrolyzed), magnesium stearate, hypromellose, titanium dioxide, Macrogol 4000, yellow ferric oxide, triacetin, and carnauba wax as excipients.

2.A.(2).2 Manufacturing process

The manufacturing process for the drug product consists of [REDACTED], packaging, and testing and storage.

[REDACTED] step have been defined as critical steps and process control items and control values have been established for [REDACTED] step.

2.A.(2).3 Control of drug product

The proposed specification for the drug product include strength, description (visual), identification (UV), uniformity of dosage units (content uniformity test), dissolution (dissolution test), and assay (HPLC). In the course of the regulatory review, the method to demonstrate uniformity of dosage units was changed from mass variation to content uniformity.

2.A.(2).4 Stability of drug product

The stability studies conducted on the drug product are as shown in Table 2. Photostability data showed that the drug product is photostable.

Table 2. Stability studies on drug product

Study	Primary batches	Temperature	Humidity	Storage package	Storage period
Long-term	3 pilot-scale batches	25°C	60%RH	PTP	24 months
Accelerated		40°C	75%RH		6 months

Based on the ICH Q1E Guideline, a shelf life of 36 months has been proposed for the drug product when packaged in PTP (polyvinyl chloride films/aluminum foils) and stored at room temperature. The long-term

stability study will continue up to 36 months.

2.B Outline of the review by PMDA

As a result of reviewing the submitted data and the responses to the questions, PMDA concluded that the quality of the drug product is adequately controlled. The key point of the review is as described below.

2.B.(1) Uniformity of dosage units

At regulatory submission, the applicant explained as follows:

istradefylline 20 mg tablets do not meet the threshold limit to allow testing uniformity by mass variation as specified in the Japanese Pharmacopoeia: “film-coated tablets, containing 25 mg or more of a drug substance comprising 25% or more, by weight, of the pre-coated tablets.” However, as the concentration relative standard deviation (RSD) of the drug substance in the dosage units is not more than █% based on data on three pilot-scale batches, the drug products may be tested for uniformity of dosage units by mass variation.

Since █ and █ to ensure the consistency of dosage units are not included in the drug product manufacturing process and an RSD of not more than █% based on data on three batches alone can not consistently guarantee a sufficiently small variability in the drug substance concentration among dosage units for future batches, PMDA instructed the applicant to test uniformity of dosage units by content uniformity instead of mass variation.

In accordance with PMDA’s instruction, the applicant included the test for uniformity of dosage units by content uniformity in the specification.

3. Non-clinical data

3.(i) Summary of pharmacology studies

3.(i).A Summary of the submitted data

3.(i).A.(1) Primary pharmacodynamics

3.(i).A.(1).1 Effects in animal models of Parkinson’s disease

3.(i).A.(1).1.(a) Reserpinized mice (Attached document 4.2.1.1-1)

i) Administration of istradefylline alone

Reserpine causes a depletion of monoamines in nerve terminals and induces parkinsonian symptoms such as transient hypokinesia and muscular rigidity (catalepsy) in rodents.

Male ddY mice were subcutaneously treated with 5 mg/kg of reserpine and those with a cataleptic score of 5 (serious) on the following day (reserpinized mice) (5 weeks of age, n = 10/group) were orally treated with istradefylline (0.01, 0.04, 0.16, 0.63, 2.5 mg/kg), L-DOPA (12.5, 25, 50, 100, 200 mg/kg), pramipexole (0.01, 0.04, 0.16, 0.63, 2.5 mg/kg), entacapone (0.63, 2.5, 10, 40, 160 mg/kg), or vehicle and then catalepsy was evaluated. Catalepsy was scored from 0 (absent) to 5 (serious) and a reversal of catalepsy was defined as a cataleptic score of ≤3. Istradefylline at ≥0.04 mg/kg, L-DOPA at ≥25 mg/kg, and pramipexole at ≥0.16 mg/kg significantly reversed reserpine-induced catalepsy compared with vehicle and the 50% effective doses (ED₅₀)

of istradefylline, L-DOPA, and pramipexole, calculated based on the number of animals in which a reversal of catalepsy was seen, were 0.19, 59, and 0.076 mg/kg, respectively. Entacapone did not reverse reserpine-induced catalepsy.

ii) Administration of istradefylline in combination with L-DOPA

The effect of istradefylline, entacapone, or vehicle in combination with L-DOPA (12.5 mg/kg) in reversing catalepsy in reserpinized mice (5 weeks of age, n = 10) was evaluated using the same procedure as the above i). L-DOPA alone did not reverse reserpine-induced catalepsy, whereas L-DOPA in combination with ≥ 0.04 mg/kg of istradefylline (0.01, 0.04, 0.16, 0.63, 2.5 mg/kg) significantly reversed reserpine-induced catalepsy compared with L-DOPA alone and the ED₅₀ of istradefylline, when combined with L-DOPA, calculated based on the number of animals in which a reversal of catalepsy was seen, was 0.019 mg/kg. L-DOPA in combination with 40 mg/kg of entacapone (0.63, 2.5, 10, 40, 160 mg/kg) significantly reversed reserpine-induced catalepsy compared with L-DOPA alone and the ED₅₀ of entacapone, when combined with L-DOPA, was 21 mg/kg.

3.(i).A.(1).1.(b) 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine-treated marmosets (Attached document 4.2.1.1-2)

i) Antiparkinsonian effect (improvement of motor symptoms)

1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine (MPTP) damages dopaminergic neurons and selectively causes a serious loss of nigral dopaminergic neurons in primates.

Male and female common marmosets (≥ 2 years of age) were treated twice at an interval of about 1 week with 2.5 mg/kg of intravenous MPTP and an additional injection of 1.5 to 2.0 mg/kg of MPTP was given to those not exhibiting overt parkinsonian symptoms at 1 week after the second injection. The animals were treated with istradefylline (0.3, 1, 3, 10 mg/kg) or vehicle if the development of parkinsonian symptoms and the antiparkinsonian effect of L-DOPA were confirmed at about 2 months after the first injection of MPTP (n = 6/sex) and then locomotor activity and motor disability scores were measured up to 8 hours post-dose. As a result, istradefylline increased locomotor activity and reduced (improved) motor disability scores in a dose-dependent manner and the effect of 10 mg/kg of istradefylline on locomotor activity and the effect of ≥ 0.3 mg/kg of istradefylline on motor disability were statistically significant compared with vehicle. The effects of istradefylline in increasing locomotor activity and improving motor disability were maintained up to at least 8 hours post-dose.

ii) Dyskinesia induction (Attached document 4.2.1.1-3)

Male and female common marmosets (≥ 2 years of age) were treated with 2.0 mg/kg of MPTP subcutaneously once daily for 5 days and about 3 weeks later, an additional injection of 2.0 mg/kg of MPTP was given. MPTP-treated marmosets were treated with 10 mg/kg of L-DOPA orally twice daily for 4 weeks to induce limb and trunk dyskinesias if the development of parkinsonian symptoms and the antiparkinsonian effect of L-DOPA were confirmed at about 2 months after the first injection of MPTP (n = 4/sex) (“marmosets previously primed to exhibit dyskinesia”). When the marmosets previously primed to exhibit

dyskinesia received vehicle and, a day later, istradefylline 10 mg/kg once daily for 21 days, the dyskinesia score after the repeated administration of istradefylline was similar to that after the administration of vehicle. Also when the animals were left untreated for 1 week after the 21-day administration and then challenged with 10 mg/kg of istradefylline, dyskinesia was not observed.

iii) Effect of istradefylline in combination with L-DOPA (Attached document 4.2.1.1-4, 4.2.1.1-5)

MPTP-treated marmosets (≥ 2 years of age, $n = 4/\text{sex}$) received a single oral dose of L-DOPA (2.5, 5.0, 7.5 mg/kg) in combination with 10 mg/kg of istradefylline or vehicle and locomotor activity and motor disability scores were measured up to 6 hours post-dose. A motor disability score (a maximal score of 17) of <9 was considered as an improvement in parkinsonian symptoms and classified as ON state and the duration of ON state (ON time) in each animal was also measured. L-DOPA dose-dependently increased locomotor activity and reduced (improved) motor disability scores. The effects of L-DOPA in combination with 10 mg/kg of istradefylline in increasing locomotor activity and improving motor disability were greater compared with the effects of L-DOPA alone at all dose levels. L-DOPA in combination with istradefylline significantly prolonged the ON time compared with L-DOPA alone at all dose levels.

iv) Effect on L-DOPA-induced dyskinesia (Attached document 4.2.1.1-6)

Marmosets previously primed to exhibit dyskinesia (≥ 2 years of age, $n = 4/\text{sex}$) were treated with 2.5 mg/kg of L-DOPA in combination with 10 mg/kg of istradefylline orally once daily for 21 days and dyskinesia scores were measured up to 6 hours post-dose. Prior to the coadministration, administration of 2.5 mg/kg of L-DOPA alone induced mild dyskinesia with a dyskinesia score of 1.50 ± 0.27 (mean \pm standard error [SE]). Following the coadministration of 2.5 mg/kg of L-DOPA and 10 mg/kg of istradefylline, the dyskinesia scores were 0.63 ± 0.18 to 1.00 ± 0.33 . Furthermore, after the 21-day administration, the animals were left untreated for 1 week and then challenged with 2.5 mg/kg of L-DOPA in combination with 10 mg/kg of istradefylline. As a result, the maximum dyskinesia score was 0.50 ± 0.19 .

3.(i).A.(1).2) Studies on the mode of action

3.(i).A.(1).2).(a) Binding affinities for adenosine receptor subtypes (Attached document 4.2.1.1-7)

Using membranes from cells expressing human or rat recombinant adenosine receptor subtypes, the inhibition constant (K_i) for the inhibition of radioligand binding by istradefylline to each of the adenosine receptor subtypes was determined. As a result, the K_i values of istradefylline for the rat and human adenosine A_{2A} receptors were 8.18 ± 0.232 and 12.4 ± 0.601 nmol/L, respectively, and the K_i value for the rat adenosine A_1 receptor was 116 ± 4.18 nmol/L. The 50% inhibitory concentration (IC_{50}) for the human adenosine A_1 receptor was >1000 nmol/L and the IC_{50} values for the rat and human adenosine A_3 receptors were both $>10,000$ nmol/L.

3.(i).A.(1).2).(b) Inhibitory activities against binding to various neurotransmitter receptors, transporters, and ion channels (Attached document 4.2.1.1-9)

Istradefylline at 10 $\mu\text{mol/L}$ inhibited the binding of specific radioligands to the adrenaline, dopamine, GABA, benzodiazepine (BZP), acetylcholine, cannabinoid, histamine, serotonin, glutamate, neurokinin, and opioid

receptors etc. and calcium channels by up to 29.59%. Although the percent inhibition of the radioligand binding to the serotonin 5-HT₇ receptor was highest at 29.59%, methiothepin as a positive control inhibited the radioligand binding to this receptor by 57.67%.

3.(i).A.(1).2).(c) MAO- and COMT-inhibitory activities (Attached document 4.2.1.1-10)

The inhibitory activities of 10 µmol/L of istradefylline against the binding of specific radioactive substrates to recombinant human monoamine oxidase (MAO)-A and MAO-B and porcine liver catechol-O-methyltransferase (COMT) were determined. As a result, istradefylline caused little inhibition of radioactive substrate binding to MAO-A or COMT and the IC₅₀ for MAO-B inhibition by istradefylline was >100,000 nmol/L.

3.(i).A.(1).2).(d) Effect on intracellular cAMP accumulation (Attached document 4.2.1.1-11)

The effect of istradefylline on an adenosine A_{2A} receptor agonist (CGS21680)-induced intracellular cAMP accumulation in PC-12 cells expressing adenosine A_{2A} receptors was studied. As a result, istradefylline (1.5-10 nmol/L) inhibited CGS21680-induced intracellular cAMP accumulation in a concentration-dependent manner and the shape of the inhibition curve indicated a competitive antagonistic effect of istradefylline. Istradefylline inhibited CGS21680-induced intracellular cAMP accumulation with a binding constant (K_B) of 0.74 ± 0.23 nmol/L. On the other hand, up to 10 nmol/L of istradefylline alone had no effect on intracellular cAMP levels.

3.(i).A.(1).2).(e) Effect on GABAergic neurotransmission in the basal ganglia (Attached document 4.2.1.1-12)

Using rat brain slices, GABA_A receptor-mediated inhibitory postsynaptic currents (IPSCs) in globus pallidus neurons were recorded using the patch-clamp method. CGS21680 at 1 µmol/L enhanced IPSCs to 145% of control (before application of CGS21680) and istradefylline at 1 µmol/L suppressed the CGS21680-induced enhancement of IPSCs to 104% of control.

3.(i).A.(1).2).(f) Effect on GABA release in the globus pallidus (Attached document 4.2.1.1-13)

Male SD/IGS rats were unilaterally lesioned by injection of 6-hydroxydopamine (6-OHDA) into the unilateral medial forebrain bundle to destroy nigral dopaminergic neurons (6-OHDA-lesioned rats) (weighing 250-340 g, n = 10) and then the extracellular levels of GABA in the globus pallidus were measured by microdialysis and the effect of istradefylline on GABA release was studied. The basal GABA levels were 9.90 ± 1.84 to 13.24 ± 2.67 nmol/L in 6-OHDA-untreated rats while they were 20.58 ± 4.11 to 25.12 ± 6.16 nmol/L in 6-OHDA-lesioned rats and pallidal GABA release increased significantly after nigrostriatal dopaminergic neuron destruction. When 6-OHDA-lesioned rats were orally treated with 1 mg/kg of istradefylline or vehicle and GABA release was measured up to 5 hours post-dose, the level of GABA release was lower in the istradefylline group than in the vehicle group.

3.(i).A.(1).3) Pharmacological potency of metabolite (Attached document 4.2.1.1.-7)

Using membranes from cells expressing human or rat recombinant adenosine receptor subtypes, the binding

affinities of istradefylline and its major metabolite in the rat and human, M1, for the adenosine A_{2A} receptor were determined in radioligand binding assays. The K_i values of istradefylline for the rat and human adenosine A_{2A} receptors were 8.18 ± 0.232 and 12.4 ± 0.601 nmol/L, respectively, and the K_i values of M1 were 6.35 ± 0.350 and 12.6 ± 0.869 nmol/L, respectively.

3.(i).A.(2) Secondary pharmacodynamics

No data have been submitted.

3.(i).A.(3) Safety pharmacology

Unless otherwise specified, safety pharmacology studies were all performed by single oral administration.

3.(i).A.(3).1 Effects on central nervous system

3.(i).A.(3).1.(a) Effects on general symptoms and behavior (Attached document 4.2.1.3-1 [Reference data], 4.2.1.3-2)

The effects of istradefylline (0.1-100 mg/kg) on general symptoms and behavior in male ddY mice (weighing 19-26 g, n = 3) were assessed by Irwin test. Istradefylline at ≥ 1 mg/kg increased stereotypy, motor activity, autonomic function, alertness, and response to stimulus and caused red skin and enlargement of the eyelids. An increase in respiratory rate was observed at ≥ 10 mg/kg.

The effects of istradefylline (10-300 mg/kg) in male CD-1 mice (6 weeks of age, n = 4) were assessed by Irwin test. At ≥ 30 mg/kg of istradefylline, increases in touch response and grooming were observed continuously after 90 minutes post-dose. Vocalization at 100 mg/kg and increased touch response at 300 mg/kg were observed from 30 minutes post-dose. The incidences of these symptoms and behaviors increased with increasing dose, but resolved by 24 hours post-dose.

3.(i).A.(3).1.(b) Spontaneous locomotor activity (Attached document 4.2.1.3-1 [Reference data], 4.2.1.3-3 [Reference data])

Male ddY mice (weighing 19-26 g, n = 5) were treated with istradefylline (0.01-300 mg/kg). An increase in spontaneous locomotor activity was observed from 90 minutes post-dose at 0.16 mg/kg and from 30 minutes post-dose at ≥ 2.5 mg/kg. The maximum increase in spontaneous locomotor activity occurred at 30 mg/kg and no further increase was observed at 100 or 300 mg/kg.

3.(i).A.(3).1.(c) Muscle relaxation and motor coordination (Attached document 4.2.1.3-1 [Reference data])

The effects of istradefylline (30-300 mg/kg) on muscle relaxation and motor coordination in male ddY mice (weighing 19-26 g, n = 5) were assessed by rotarod, inclined screen, and traction tests. Istradefylline had no effect on muscle relaxation or motor coordination at any dose level.

3.(i).A.(3).1.(d) Pentobarbital-induced sleeping time (Attached document 4.2.1.3-1 [Reference data])

Male ddY mice (weighing 19-26 g, n = 10) were treated with istradefylline (0.1-300 mg/kg). Istradefylline at ≥ 10 mg/kg reduced pentobarbital-induced sleeping time.

3.(i).A.(3).1).(e) Anti- or pro-convulsant effects (Attached document 4.2.1.3-1 [Reference data], 4.2.1.3-6 [Reference data])

Male ddY mice (weighing 19-26 g, n = 5-10) were treated with istradefylline (1-300 mg/kg). Istradefylline had no effect on pentylenetetrazol- or electroshock-induced seizures at any dose level.

3.(i).A.(3).1).(f) Body temperature (Attached document 4.2.1.3-1 [Reference data], 4.2.1.3-7 [Reference data], 4.2.1.3-9 [Reference data], 4.2.1.3-10)

Male ddY mice (weighing 19-26 g, n = 10) were treated with istradefylline (0.01-100 mg/kg). Istradefylline produced a maximum rise in body temperature of 1.5°C above that of the vehicle group (30 minutes after administration of 10 mg/kg of istradefylline). There were no effects on body temperature at ≤ 0.1 mg/kg.

Male albino hybrid rabbits (weighing 2.5-3.2 kg, n = 5-6) were treated with istradefylline (0.03-300 mg/kg). Body temperature rose by 0.7°C (5.5 hours post-dose) in the 300 mg/kg group compared with that in the vehicle group. There were no effects on body temperature at ≤ 30 mg/kg.

Male beagle dogs (14 months of age, n = 4) were treated with istradefylline (8-400 mg/kg). Istradefylline had no effect on body temperature at any dose level.

3.(i).A.(3).1).(g) Analgesia (Attached document 4.2.1.3-1 [Reference data])

Male ddY mice (weighing 19-26 g, n = 7-10) were treated with istradefylline (1-300 mg/kg). Istradefylline had no effect on the number of writhes induced by intraperitoneal 0.7% acetic acid or the reaction time to the pain induced by a pressure applied at the root of the tail at any dose level.

3.(i).A.(3).1).(h) Electroencephalogram (EEG) (Attached document 4.2.1.3-11 [Reference data], 4.2.1.3-12 [Reference data])

Male SD rats (weighing 220-280 g, n = 6) were treated with istradefylline (0.1-300 mg/kg). Istradefylline had no effect on spontaneous EEG at any dose level.

3.(i).A.(3).1).(i) Conditioned avoidance behavior (Attached document 4.2.1.3-13 [Reference data])

Male SD rats (weighing 220-280 g, n = 10) were treated with istradefylline (0.03-300 mg/kg). A reduction in the reaction time at ≥ 0.3 mg/kg, an increase in the avoidance rate at 3 mg/kg, and an increase in the number of shuttles at 30 mg/kg were observed.

3.(i).A.(3).2) Effects on cardiovascular and respiratory systems

3.(i).A.(3).2).(a) hERG currents (Attached document 4.2.1.3-14)

Istradefylline (0.1-2 $\mu\text{mol/L}$ [measured concentrations, 0.0729-1.71 $\mu\text{mol/L}$]) was applied to HEK293 cells expressing human ether-a-go-go-related gene (hERG) channels. Istradefylline had no effect on hERG currents at any concentration.

3.(i).A.(3).2).(b) Effects on cardiovascular and respiratory systems in the dog and rat (Attached document 4.2.1.3-10, 4.2.1.3-15 [Reference data])

Male beagle dogs (14 months of age, n = 4) were treated with istradefylline (8-400 mg/kg). Significant increases in diastolic blood pressure and mean blood pressure were sporadically observed up to 8 hours post-dose at all dose levels of istradefylline compared with vehicle. An increase in heart rate was observed at 6 hours post-dose with 400 mg/kg of istradefylline compared with vehicle. The significant increases in blood pressure and heart rate were considered associated with decreases from baseline in the vehicle group. Istradefylline had no effect on other cardiovascular (systolic blood pressure, ECG) or respiratory (respiratory rate, blood gases) parameters.

Male SD rats (weighing 270-330 g, n = 6) were treated with istradefylline (0.3-10 mg/kg). Slight increases in blood pressure and heart rate were observed at all dose levels. The increases in blood pressure and heart rate were similar across all dose groups.

3.(i).A.(3).2).(c) Incidence of arrhythmias in dog model of myocardial infarction (Attached document 4.2.1.3-16 [Reference data], 4.2.1.3-17, 4.2.1.3-18 [Reference data])

Myocardial infarction model was developed in male beagle dogs (15-25 months of age, n = 6-12) by partial ligation of the left anterior descending coronary artery. Istradefylline 10 or 30 mg/kg was administered 2 days after the induction of myocardial infarction and ECG was measured up to 10 hours post-dose. Ventricular fibrillation did not occur and no effects on sustained ventricular tachycardia (VTs) were observed at either dose level. The incidence of non-sustained ventricular tachycardia (VTn) was unaffected at 30 mg/kg while it was significantly higher at 10 mg/kg compared with vehicle. The incidence of VTn at baseline was also significantly higher in the 10 mg/kg group than in the vehicle group.

3.(i).A.(3).2).(d) Coronary reactive hyperemia (Attached document 4.2.1.3-21, 4.2.1.3-22 [Reference data])

Male beagle dogs (10-16 months of age, n = 6) were treated with 40 to 400 mg/kg of istradefylline orally once daily for 5 days and the effect of istradefylline on reactive hyperemia induced by ligation and reperfusion of the coronary artery was assessed. Istradefylline had no effect on reactive hyperemia or cardiac hemodynamics (mean blood pressure, heart rate, cardiac output, left ventricular pressure, left ventricular end-diastolic pressure, maximum rate of increase of left ventricular pressure) at any dose level. Ventricular fibrillation or ventricular tachycardia was not induced under any condition.

3.(i).A.(3).3) Effects on gastrointestinal system

3.(i).A.(3).3).(a) Small intestinal transport (Attached document 4.2.1.3-1 [Reference data])

The effect of istradefylline (10-300 mg/kg) on charcoal transit in the small intestine of male ddY mice (weighing 19-26 g, n = 10) was assessed. Istradefylline had no effect on small intestinal transport at any dose level.

3.(i).A.(3).3.(b) Small intestinal transport and gastric emptying following administration of istradefylline in combination with levodopa/carbidopa (Attached document 4.2.1.3-25, 4.2.1.3-26)

The effects of 100 mg/kg of istradefylline alone and 100 mg/kg of istradefylline in combination with levodopa/carbidopa (250/25 mg/kg) on charcoal transit in the small intestine of male SD rats (5 weeks of age, n = 6) were assessed. As a result, istradefylline, either alone or in combination with levodopa/carbidopa, had no effect on small intestinal transport.

The effects of 100 mg/kg of istradefylline alone, levodopa/carbidopa (250/25 mg/kg) alone, and 100 mg/kg of istradefylline in combination with levodopa/carbidopa (250/25 mg/kg) on the gastric emptying of male SD rats (5 weeks of age, n = 6) were assessed using the phenol red method. The percent gastric emptying was $74.1 \pm 9.1\%$ in the vehicle group, $96.5 \pm 1.8\%$ in the istradefylline alone group, $19.5 \pm 6.2\%$ in the levodopa/carbidopa alone group, and $9.8 \pm 2.4\%$ in the istradefylline plus levodopa/carbidopa group and istradefylline alone had no effect on gastric emptying. While levodopa inhibited gastric emptying, istradefylline did not affect levodopa-induced inhibition of gastric emptying.

3.(i).A.(4) Pharmacodynamic drug interactions

No data have been submitted.

3.(i).B Outline of the review by PMDA

3.(i).B.(1) Antiparkinsonian effects

The applicant explained the antiparkinsonian effects of istradefylline as follows:

Istradefylline has been shown to be a selective adenosine A_{2A} receptor antagonist since it exhibited a high binding affinity for the adenosine A_{2A} receptor, did not affect other neurotransmitter receptors, transporters, or MAO or COMT involved in the metabolism of dopamine, and inhibited an adenosine A_{2A} receptor agonist-induced intracellular cAMP accumulation. In various animal models of Parkinson's disease (reserpinized mice, MPTP-treated marmosets), istradefylline alone demonstrated antiparkinsonian effects and istradefylline in combination with L-DOPA increased the antiparkinsonian activity compared with either drug alone. Furthermore, istradefylline at doses producing antiparkinsonian effects did not induce dyskinesia or exacerbate L-DOPA-induced dyskinesia and did not cause dopaminergic symptoms such as vomiting, seizures, and stereotypy. On the other hand, istradefylline reduced the activity of the indirect striato-pallidal pathway, which is considered to play an important role in the pathogenesis of Parkinson's disease. The above findings indicate that istradefylline produces antiparkinsonian effects by acting through a different mechanism from conventional anti-Parkinson's medications and istradefylline is expected to show antiparkinsonian activity without dopaminergic side effects and as adenosine A_{2A} receptor-mediated modulation of the activity of GABAergic neurons of the indirect striato-pallidal pathway is independent of dopaminergic modulation of this pathway, istradefylline in combination with other anti-Parkinson's medications such as L-DOPA is expected to increase the antiparkinsonian activity.

In a non-clinical study using rat brain slices, the concentration of istradefylline that reduced the activity of the indirect striato-pallidal pathway (1 $\mu\text{mol/L}$) was higher than the K_i for the adenosine A_{2A} receptor (12.4 nmol/L). PMDA asked the applicant to explain whether a similar action can be expected also in humans at clinical doses.

The applicant responded as follows:

Istradefylline concentrations exerting its action should depend on the concentration of an adenosine receptor agonist, CGS21680, used in the experimental system (1 $\mu\text{mol/L}$) or the K_i (19-27 nmol/L) (Fredholm BB et al. *Pharmacol Rev.* 2011;63:1-34.). In light of the K_i of istradefylline for the adenosine A_{2A} receptor (12.4 nmol/L), 1 $\mu\text{mol/L}$ as the concentration of istradefylline required to antagonize 1 $\mu\text{mol/L}$ of CGS21680 is largely appropriate. As the K_i of adenosine for the human adenosine A_{2A} receptor is around 310 nmol/L (Fredholm BB et al. *Pharmacol Rev.* 63: 1-34, 2011), the K_i of istradefylline (12.4 nmol/L) shows that istradefylline can fully antagonize the action of endogenous adenosine. Following 14-day oral administration of 20 and 40 mg/day of istradefylline to humans, the plasma trough concentrations at steady state were 154.6 and 284.7 ng/mL, respectively, and given that the protein binding of istradefylline is 97% in humans, the plasma trough unbound concentrations are calculated to be 12.1 and 22.2 nmol/L, respectively. Therefore, following administration of clinical doses of istradefylline in humans, plasma unbound concentrations at steady state will be higher than the K_i for the adenosine A_{2A} receptor and istradefylline is expected to exert a similar antagonistic action on endogenous adenosine modulation of GABAergic neurotransmission in the basal ganglia via the adenosine A_{2A} receptor.

PMDA considers as follows:

Since *in vitro* studies demonstrated that istradefylline is a selective adenosine A_{2A} receptor antagonist and dose-dependent antiparkinsonian effects of istradefylline were observed in different animal models of Parkinson's disease, the antiparkinsonian efficacy of istradefylline can be expected. Whether clinically used istradefylline produces antiparkinsonian effects by antagonizing adenosine A_{2A} receptor-mediated modulation of GABAergic neurotransmission in the basal ganglia is not necessarily clear. However, as istradefylline in combination with L-DOPA increased the antiparkinsonian activity in different animal models of Parkinson's disease, the efficacy of istradefylline in combination with levodopa preparations can be expected also in patients with Parkinson's disease.

3.(i).B.(2) Safety pharmacology studies

PMDA asked the applicant to discuss the observed effect of istradefylline on the occurrence of arrhythmias in a dog model of myocardial infarction and explain the potential for clinical doses of istradefylline to exacerbate arrhythmias in humans.

The applicant responded as follows:

Generally, a large amount of adenosine formed from ATP degradation and released from the myocardium during myocardial ischemia is considered to exert cardioprotective effects by reducing heart rate via the adenosine A_1 receptor and dilating coronary vessels and inhibiting neutrophil activation etc. via the

adenosine A_{2A} receptor (Peart JN et al. *Pharmacol Ther.* 2007;114: 208-21). Therefore, it was thought that the adenosine A_{2A} receptor antagonism of istradefylline might exacerbate arrhythmias induced by myocardial ischemia and the potential for istradefylline to exacerbate arrhythmias was examined using a dog model of acute myocardial infarction. As a result, although the possibility that 30 mg/kg of istradefylline exacerbated arrhythmias during acute myocardial infarction was suggested, as the conclusion of the study using dogs with acute myocardial infarction was questionable [REDACTED], the report was reevaluated. As a result, the method of analysis was considered inappropriate because ectopic beat classification was not in accordance with the Lambeth conventions, the global standard guidelines (Walker MJA et al. *Cardiovas Res.* 1988;22:447-55) and between-group comparison was not performed etc. Thus, taking account of the above, a reanalysis was performed. As a result, ventricular fibrillation did not occur and no effects on sustained ventricular tachycardia (VTs) or non-sustained ventricular tachycardia (VTn) were observed at either dose level. Thus, it was concluded that istradefylline does not exacerbate arrhythmias in this model. In the study using dogs with acute myocardial infarction, the plasma concentrations of unbound istradefylline were 22.1 ng/mL (2 hours post-dose) to 19.4 ng/mL (8 hours post-dose), which were almost equal to the C_{max} of unbound istradefylline following administration of 40 mg/day in humans (18.4 ng/mL). Therefore, at least within the clinical effective plasma concentration range, istradefylline is unlikely to induce arrhythmia. Also in a dog model of reactive hyperemia, istradefylline had no effect on reactive hyperemia induced by brief ischemia and did not induce arrhythmia. Thus, istradefylline is unlikely to exacerbate recovery from ischemia. In Japanese clinical studies of istradefylline in patients with Parkinson's disease, ECG changes or increased arrhythmias were not observed and as adverse drug reactions classified as ECG abnormalities, supraventricular extrasystoles [0.6% (4 of 649 subjects)], palpitations [0.5% (3 of 649 subjects)], atrial fibrillation [0.5% (3 of 649 subjects)], electrocardiogram T wave inversion [0.3% (2 of 649 subjects)], ventricular extrasystoles [0.3% (2 of 649 subjects)], electrocardiogram ST segment depression [0.2% (1 of 649 subjects)], and supraventricular tachycardia [0.2% (1 of 649 subjects)] were reported, but all events were mild or moderate in severity and none were reported as serious adverse events. Thus, these events were not considered clinically relevant.

PMDA considers as follows:

The applicant discussed that based on the results of a reanalysis of data from a non-clinical study using a dog model of myocardial infarction, istradefylline is unlikely to induce arrhythmia within the clinical effective plasma concentration range. However, as it is difficult to say that the baseline was appropriately controlled in this non-clinical pharmacology study, the possibility of exacerbation of arrhythmias during acute myocardial infarction, as expected from the mode of action of istradefylline, can not be excluded, based only on this study. Although Japanese clinical studies did not clearly indicate the risk of ECG changes or increased arrhythmias, since patients with clinically significant illness of any organ system, e.g. the heart (including serious abnormal findings in the ECG at enrollment) were excluded from the Japanese clinical studies, it is necessary to caution about the risk of exacerbation of arrhythmias by istradefylline during myocardial ischemia. The specific caution statement will be finalized taking account of comments from the Expert Discussion.

3.(ii) Summary of pharmacokinetic studies

3.(ii).A Summary of the submitted data

Plasma concentrations of istradefylline were determined by validated high performance liquid chromatography-ultraviolet detection (HPLC-UV) and liquid chromatography/tandem mass spectrometry (LC/MS/MS) methods. The lower limit of quantitation of the HPLC-UV method was 5 ng/mL in rat and dog plasma and the lower limit of quantitation of the LC/MS/MS method was 1 ng/mL in mouse, rat, and dog plasma. Radioactivity in samples was measured by liquid scintillation counter. In studies to determine the plasma, urinary, and fecal metabolite profiles, samples were fractionated by high performance liquid chromatography (HPLC) and then radioactivity was measured by liquid scintillation counter or solid scintillation counter. Unless otherwise specified, pharmacokinetic parameters are expressed as the mean.

3.(ii).A.(1) Absorption

3.(ii).A.(1).1 Single-dose pharmacokinetics (Attached document 4.2.2.2-2 to 4.2.2.2-6)

Following single oral doses of 0.3, 3, and 30 mg/kg of istradefylline in male mice ($n = 5/\text{group}$), the time to reach the maximum plasma concentration (t_{\max}) values of istradefylline were 0.5, 1.0, and 5.0 hours, respectively, the maximum plasma concentration (C_{\max}) values were 32.4, 493.2, and 4270.0 ng/mL, respectively, and the area under the plasma concentration-time curve up to 24 hours (AUC_{0-24}) values were 195, 2478, and 50,971 ng·h/mL, respectively.

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male rats ($n = 4$), the t_{\max} of plasma radioactivity was 2.0 hours, the C_{\max} was 449.0 ng eq./mL, the area under the plasma concentration-time curve from zero to infinity ($AUC_{0-\infty}$) was 5620 ng eq·h/mL, and the plasma elimination half-life ($t_{1/2}$) was 7.31 hours. Following single oral doses of 0.3, 3, 30, and 300 mg/kg of istradefylline in male rats ($n = 4/\text{group}$), the t_{\max} values of istradefylline were 2.0, 3.0, 4.0, and 6.5 hours, respectively, the C_{\max} values were 14.8, 176, 859, and 2060 ng/mL, respectively, and the $AUC_{0-\infty}$ values were 100, 1460, 9230, and 33,700 ng·h/mL, respectively. The $t_{1/2}$ values in the 0.3, 3, and 30 mg/kg groups were 3.32, 3.26, and 4.44 hours, respectively, and in the istradefylline 300 mg/kg group, istradefylline in plasma was eliminated in a biphasic manner, with an alpha-phase $t_{1/2}$ of 5.56 hours and a beta-phase $t_{1/2}$ of 37.6 hours. Following a single intravenous dose of 0.3 mg/kg of istradefylline in male rats ($n = 4$), the $AUC_{0-\infty}$ of istradefylline was 292 ng·h/mL, the $t_{1/2}$ was 3.49 hours, total body clearance (CL) was 1.05 L/h/kg, and the steady-state volume of distribution (V_{dss}) was 3.44 L/kg. The absolute bioavailability (BA) of istradefylline after oral administration of 0.3 mg/kg of istradefylline to male rats was 34.3%.

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male dogs ($n = 3$), the t_{\max} of plasma radioactivity was 0.8 hours, the C_{\max} was 506.6 ng eq./mL, and the $AUC_{0-\infty}$ was 5906 ng eq·h/mL. Radioactivity in plasma was eliminated triphasically and the terminal phase $t_{1/2}$ was 148.05 hours. Following a single intravenous dose of 0.3 mg/kg of istradefylline in male dogs ($n = 3$), the $AUC_{0-\infty}$ of istradefylline was 281 ng·h/mL, the $t_{1/2}$ was 6.69 hours, the CL was 1.13 L/h/kg, and the V_{dss} was 6.60 L/kg. Following single oral doses of 0.3, 3, 30, and 300 mg/kg of istradefylline in fasted male dogs ($n = 3/\text{group}$), the t_{\max}

values of istradefylline were 1.0, 1.0, 1.2, and 12 hours, respectively, the C_{\max} values were 15.9, 80.0, 221, and 793 ng/mL, respectively, the $AUC_{0-\infty}$ values were 90.7, 454, 1930, and 16,700 ng·h/mL, respectively, and the $t_{1/2}$ values were 4.78, 5.76, 8.39, and 8.85 hours, respectively. The absolute BA of istradefylline after oral administration of 0.3 mg/kg of istradefylline to fasted male dogs was 30.5%. Following single oral doses of 0.3, 3, 30, and 300 mg/kg of istradefylline in fed male dogs ($n = 3/\text{group}$), the t_{\max} values of istradefylline were 1.0, 1.3, 3.3, and 8.0 hours, respectively, the C_{\max} values were 15.2, 148, 1150, and 3240 ng/mL, respectively, the $AUC_{0-\infty}$ values were 119, 1400, 17,600, and 80,400 ng·h/mL, respectively, and the $t_{1/2}$ values were 7.51, 5.88, 7.57, and 9.18 hours, respectively, and the absolute BA of istradefylline after oral administration of 0.3 mg/kg of istradefylline under fed conditions was 42.3%.

3.(ii).A.(1).2) Repeat-dose pharmacokinetics (Attached document 4.2.3.2-3, 4.2.3.2-18)

Istradefylline at doses of 6, 30, 160, and 800 mg/kg was orally administered once daily for 28 days to male and female rats ($n = 3/\text{group}$). The C_{\max} and AUC_{0-24} on Days 1 and 28 increased with increasing dose, but the increases in the C_{\max} and AUC_{0-24} were less than dose-proportional. No marked gender-related differences were observed for the C_{\max} and AUC_{0-24} on Days 1 and 28 at any dose level. There were no marked increases in the C_{\max} or AUC_{0-24} on Day 28 compared to Day 1.

Istradefylline at doses of 25, 100, and 400 mg/kg was orally administered once daily for 28 days to male and female dogs ($n = 4/\text{group}$) and plasma concentrations were measured on Days 1 and 23. The C_{\max} and AUC_{0-24} both increased with increasing dose, but the increases in the C_{\max} and AUC_{0-24} were less than dose-proportional. No marked gender-related differences were observed for the C_{\max} and AUC_{0-24} on Days 1 and 23 at any dose level. The C_{\max} and AUC_{0-24} values on Day 23 were higher than those on Day 1, with about 1.4- to 4.0-fold and 1.7- to 5.1-fold increases, respectively, and there was no clear relationship between the dose level and the increases after repeated administration.

3.(ii).A.(2) Distribution

3.(ii).A.(2).1) Tissue distribution (Attached document 4.2.2.3-1)

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male albino rats ($n = 4/\text{timepoint}$), tissue radioactivity levels were determined at 2, 6, and 48 hours post-dose. Radioactivity levels peaked at 6 hours post-dose in the small and large intestines and at 2 hours post-dose in other tissues. The peak level of radioactivity per gram of tissue was highest in the small intestine, followed in decreasing order by white fat, large intestine, brown fat, liver, adrenal gland, Harderian gland, stomach, kidneys, pancreas, mesenteric lymph nodes, and thyroid gland. When the peak level of radioactivity per gram of tissue was compared with the peak level of radioactivity per mL of plasma, the peak radioactivity levels in the small intestine, white fat, large intestine, brown fat, and liver were approximately 42-, 22-, 16-, 13-, and 8-fold the peak radioactivity level in plasma, respectively, and the peak radioactivity levels in other tissues were approximately 2- to 6-fold the peak radioactivity level in plasma. The radioactivity levels in the brain were 1.26- and 0.99-fold the plasma radioactivity levels at 2 and 6 hours post-dose, respectively, and the radioactivity level in the brain was less than half the plasma level at 48 hours post-dose. Radioactivity in most tissues was eliminated in parallel with plasma radioactivity and the radioactivity levels in the large

intestine, thyroid gland, and other tissues at 48 hours post-dose were about 1/3, 1/9, and $\leq 1/15$ of those at 2 hours post-dose, respectively.

Although no tissue distribution studies in pigmented rats have been performed, a phototoxicity study in pigmented rats has been conducted [see “3.(iii) Summary of toxicology studies”].

3.(ii).A.(2).2) Protein binding (Attached document 4.2.2.3-2 to 4.2.2.3-4, 4.2.2.3-6)

When ^{14}C -istradefylline was added to rat serum at final concentrations of 10, 100, 1000, and 10,000 ng/mL, the percent protein binding values were 95.6% to 96.1%. Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male rats ($n = 3/\text{timepoint}$), the percent serum protein binding values at 0.5, 2, and 6 hours post-dose were 95.1%, 94.1%, and 92.1%, respectively.

When ^{14}C -istradefylline was added to dog serum at final concentrations of 10, 100, 1000, and 10,000 ng/mL, the percent protein binding values were 90.9% to 92.5%. Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male dogs ($n = 3$), the percent plasma protein binding values at 0.5, 2, and 8 hours post-dose were 94.0%, 91.8%, and 90.7%, respectively.

When ^{14}C -istradefylline was added to rabbit serum at final concentrations of 10, 100, 1000, and 10,000 ng/mL, the percent protein binding values were 97.8% to 98.1%.

3.(ii).A.(2).3) Distribution in blood cells (Attached document 4.2.2.2-3, 4.2.2.2-5)

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male rats ($n = 4$), the blood-to-plasma ratio of total radioactivity was 0.61 to 0.74 at 0.25 to 36 hours post-dose.

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male dogs ($n = 3$), the blood-to-plasma ratio of total radioactivity was 0.65 to 0.89 at 0.25 to 48 hours post-dose.

3.(ii).A.(2).4) Placental transfer (Attached document 4.2.2.3-7)

Rats on gestation days 12 and 19 ($n = 3/\text{timepoint}$) were treated with a single oral dose of 3 mg/kg of ^{14}C -istradefylline and radioactivity levels in maternal tissues and fetuses were measured at 2, 8, and 24 hours post-dose for rats on gestation day 12 and maternal and fetal tissue radioactivity levels were measured at 2, 8, and 24 hours post-dose for rats on gestation day 19. In rats on gestation day 12, radioactivity levels in maternal tissues excluding white fat and in fetuses peaked at 2 hours post-dose and declined to $\leq 15\%$ of the peak levels at 24 hours post-dose. The fetal-to-maternal blood ratio of radioactivity at 2 hours post-dose was 0.7. In rats on gestation day 19, fetal radioactivity levels peaked at 2 hours post-dose and declined to about 52.6% of the peak levels at 24 hours post-dose. The fetal-to-maternal blood ratio of radioactivity was ≥ 1 at all timepoints.

3.(ii).A.(3) Metabolism

3.(ii).A.(3).1) *In vitro* metabolism (Attached document 4.2.2.4-7)

Male and female rat hepatocytes were incubated with 10 µmol/L of istradefylline at 37°C for 4 hours. In both male and female rat hepatocytes, three different glucuronide conjugates of demethyl istradefylline, a sulfate conjugate of demethyl istradefylline, 1-deethyl istradefylline, 3-deethyl istradefylline, M8 (1-β-hydroxylated istradefylline), *cis*-istradefylline, and M1 (4'-*O*-demethyl istradefylline) were detected. Male and female rat hepatocytes were incubated with 10 µmol/L of ¹⁴C-istradefylline at 37°C for 4 hours. In both male and female rat hepatocytes, ≥90% remained as unchanged drug and M1 was the predominant metabolite.

When female rabbit hepatocytes were incubated with 10 µmol/L of istradefylline at 37°C for 4 hours, two different glucuronide conjugates of demethyl istradefylline, a sulfate conjugate of demethyl istradefylline, 1-deethyl istradefylline, 3-deethyl istradefylline, M8, *cis*-istradefylline, and M1 were detected. When female rabbit hepatocytes were incubated with 10 µmol/L of ¹⁴C-istradefylline at 37°C for 4 hours, 48.7% remained as unchanged drug and glucuronide conjugates of demethyl istradefylline accounted for approximately 80% of the metabolites formed.

3.(ii).A.(3).2 *In vivo* metabolism (Attached document 4.2.2.4-1 to 4.2.2.4-6)

Following a single oral dose of 3 mg/kg of ¹⁴C-istradefylline in male mice (n = 6/timepoint), metabolites in plasma at 1, 2, and 6 hours post-dose were determined. Istradefylline accounted for 44.73% to 55.13% of the total radioactivity in plasma and M5 (a glucuronide conjugate of M1) accounted for 35.92% to 41.95% of the total radioactivity in plasma. M1, M8, and M4 (a sulfate conjugate of M1) accounted for 3.83% to 4.45%, 0.56% to 2.17%, and 0.64% of the total radioactivity in plasma, respectively.

Following a single oral dose of 3 mg/kg of ¹⁴C-istradefylline in male rats (n = 3/timepoint), metabolites in plasma at 2 and 6 hours post-dose were determined. Istradefylline, M5, M1, M4, and M8 were present in plasma and accounted for 45.03%, 17.39%, 7.78%, 1.41%, and 0.90%, respectively, of the total radioactivity at 6 hours post-dose. Glucuronide conjugates of demethyl istradefylline were also detected.

Following a single oral dose of 3 mg/kg of ¹⁴C-istradefylline in male rats (n = 3), 17.6% of the administered radioactivity was excreted in urine up to 48 hours post-dose and the major metabolites excreted in urine were M3 (3',4'-*O*-didemethyl istradefylline) and M1, which represented 5.31% and 1.96% of the administered radioactivity, respectively. Istradefylline was not detected in urine. Also, 68.3% of the administered radioactivity was excreted in feces up to 48 hours post-dose and the major metabolites excreted in feces were M3, M1, and M10 (hydrogenated M3), which represented 30.60%, 9.34%, and 8.33% of the administered radioactivity, respectively, and 1.62% of the administered radioactivity was excreted in feces as istradefylline.

Following a single oral dose of 3 mg/kg of ¹⁴C-istradefylline in bile duct cannulated male rats (n = 3), 86.8% of the administered radioactivity was excreted in bile up to 24 hours post-dose. The major metabolites in bile were M5, M4, and M1, which represented 40.5%, 31.4%, and 4.00% of the administered radioactivity, respectively. Istradefylline was not detected in bile.

Following a single oral dose of 3 mg/kg of ¹⁴C-istradefylline in male rats (n = 3/timepoint), metabolites in

plasma and brain at 0.5, 2, and 6 hours post-dose were determined. In the brain, istradefylline was predominant, followed by M1 and istradefylline and M1 accounted for 79.5% to 85.7% and 8.39% to 16.3% of the total radioactivity in the brain, respectively. The brain-to-plasma ratios of radioactivity for istradefylline and M1 were 1.69 to 2.21 and 1.42 to 2.85, respectively.

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male dogs ($n = 3$), istradefylline, M4, M5, M8, and M1 were primarily present in plasma and accounted for 43.82% to 74.49%, 9.84% to 18.66%, 1.91% to 13.11%, 1.94% to 4.50%, and 0.79% to 2.40%, respectively, of the total radioactivity in plasma. Up to 48 hours post-dose, 4.5% of the administered radioactivity was excreted in urine and 83.3% in feces and 0.08% of the administered radioactivity was excreted in urine as istradefylline and 28.25% in feces as istradefylline. The major metabolite in urine was M4 (1.38% of the administered radioactivity) and the major metabolite in feces was M10 (26.36% of the administered radioactivity).

3.(ii).A.(4) Excretion

3.(ii).A.(4).1 Urinary and fecal excretion (Attached document 4.2.2.2-3, 4.2.2.2-5)

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male rats ($n = 4$), 21.2% and 78.4% of the administered radioactivity were excreted in urine and feces, respectively, up to 168 hours post-dose.

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in male dogs ($n = 3$), 6.8% and 92.7% of the administered radioactivity were excreted in urine and feces, respectively, up to 168 hours post-dose.

3.(ii).A.(4).2 Biliary excretion and enterohepatic circulation (Attached document 4.2.2.5-1 to 4.2.2.5-2)

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in bile duct cannulated male rats ($n = 5$), 77.5%, 6.9%, and 12.6% of the administered radioactivity were excreted in bile, urine, and feces, respectively, up to 48 hours post-dose.

When bile collected after a single oral dose of 3 mg/kg of ^{14}C -istradefylline from bile duct cannulated donor male rats was intraduodenally administered to bile duct cannulated recipient male rats ($n = 3$), 75.8%, 7.3%, and 17.3% of the administered radioactivity were excreted in bile, urine, and feces, respectively, up to 48 hours post-dose.

3.(ii).A.(4).3 Excretion in milk (Attached document 4.2.2.5-3)

Following a single oral dose of 3 mg/kg of ^{14}C -istradefylline in lactating rats on lactation day 9 ($n = 3$), the t_{max} values of radioactivity in blood and milk were 2.8 and 1.0 hours, respectively, the C_{max} values were 419 and 4590 ng eq./mL, respectively, and the AUC_{0-t} values were 4580 and 30,100 ng eq.·h/mL, respectively. Radioactivity was detected also in blood from suckling rats and blood radioactivity levels peaked at 79 ng eq./mL at 12 hours after administration of ^{14}C -istradefylline to lactating rats and then declined slowly.

3.(ii).A.(5) Pharmacokinetic drug interactions with levodopa/carbidopa (Attached document 4.2.2.6-1

to 4.2.2.6-3)

Following a single oral dose of 1, 3, or 100 mg/kg of istradefylline alone or in combination with levodopa/carbidopa (250/25 mg/kg) in male rats ($n = 3/\text{group}$), the ratio of combination therapy to monotherapy for the area under the plasma concentration-time curve from zero to the last sampling point (AUC_{0-t}) of istradefylline increased with increasing dose, i.e. 1.05, 1.19, and 1.34, respectively, whereas there were no differences in the AUC_{0-t} ratio of M1 to istradefylline between monotherapy and combination therapy.

Male rats ($n = 3/\text{group}$) were treated with a single intravenous dose of 0.3 mg/kg of istradefylline alone or with oral levodopa/carbidopa (250/25 mg/kg) followed 30 minutes later by a single intravenous dose of 0.3 mg/kg of istradefylline. The $\text{AUC}_{0-\infty}$ values of plasma istradefylline were 403 and 334 ng·h/mL, respectively, the $t_{1/2}$ values were 2.70 and 2.75 hours, respectively, the CL values were 0.762 and 0.900 L/h/kg, respectively, and the V_{dss} values were 2.59 and 2.88 L/kg, respectively, and the t_{max} values of plasma M1 were 1.00 and 0.56 hours, respectively, the C_{max} values were 8.41 and 7.74 ng/mL, respectively, and the AUC_{0-t} values were 30.6 and 28.9 ng·h/mL, respectively.

At 24 hours after a single oral dose of 1 mg/kg of ^{14}C -istradefylline alone or in combination with levodopa/carbidopa (250/25 mg/kg) in male rats ($n = 3$), 14.9% and 17.1%, respectively, of the administered radioactivity were excreted in urine, 16.1% and 15.7%, respectively, of the administered radioactivity were excreted in feces, and 5.42% and 7.66%, respectively, of the administered radioactivity were retained in the body. At 24 hours after a single oral dose of 100 mg/kg of ^{14}C -istradefylline alone or in combination with levodopa/carbidopa in male rats ($n = 3$), 7.07% and 12.4%, respectively, of the administered radioactivity were excreted in urine, 55.7% and 28.2%, respectively, of the administered radioactivity were excreted in feces, and 6.03% and 21.1%, respectively, of the administered radioactivity were retained in the body.

3.(ii).B Outline of the review by PMDA

3.(ii).B.(1) Food effect

When single oral doses of istradefylline were administered to dogs under fed conditions, the plasma concentrations of istradefylline were increased compared with administration under fasted conditions. PMDA asked the applicant to explain the cause for greater food effects with higher doses.

The applicant explained as follows:

Following single oral doses of 0.3 to 300 mg/kg of istradefylline in fasted male dogs, the C_{max} and $\text{AUC}_{0-\infty}$ increased with increasing dose, but the increases in the C_{max} and $\text{AUC}_{0-\infty}$ were less than dose-proportional. Since istradefylline is a drug of high membrane permeability, with a low solubility in water (0.6 µg/mL), it is inferred that the dissolution of higher doses of istradefylline in the gastrointestinal tract was limited, leading to a reduced absorption rate. On the other hand, when the same single oral doses of istradefylline were administered to fed male dogs, the exposures at higher doses of ≥ 3 mg/kg were increased compared with administration under fasted conditions and the increases in the C_{max} and $\text{AUC}_{0-\infty}$ were largely dose-proportional over the dose range of 0.3 to 30 mg/kg. Some insoluble drugs become more soluble and thus

available for absorption when bile secretion is increased by a meal. It is inferred that when administered with food, istradefylline also became more soluble in the gastrointestinal tract and the rate of absorption was maintained even at higher doses of ≥ 3 mg/kg, resulting in an increased exposure.

PMDA's view on the food effect on the pharmacokinetics of istradefylline is as follows:

Non-clinical studies in dogs showed that the plasma concentrations of istradefylline were increased in fed than in fasted dogs and that the increases in the plasma concentration became more prominent with higher doses. It seems that the food effect differed depending on the dose of istradefylline in the non-clinical studies in dogs because of differences in the dissolution of istradefylline in the gastrointestinal tract. In humans, the exposure of istradefylline increased dose-proportionally over the clinical dose range and it has been demonstrated that the food effect does not significantly differ across the doses when istradefylline is administered at doses ranging from 20 to 50 mg [see "4.(i) Summary of biopharmaceutic studies and associated analytical methods" and "4.(ii) Summary of clinical pharmacology studies"]. Therefore, when istradefylline is administered in the clinical dose range to humans, the food effect on the pharmacokinetics of istradefylline should not differ across the doses. When istradefylline was administered to humans after a meal, the exposure was increased by 10% to 20% compared with administration under fasted conditions, and the efficacy and safety of istradefylline have been confirmed in a phase III study (Study 6002-009) and a long-term treatment study (Study 6002-010) where istradefylline was administered without regard to meals. Therefore, there is no need to specify the timing of dosing relative to meals in the dosage and administration section of the package insert for istradefylline.

3.(ii).B.(2) Istradefylline interactions with levodopa/carbidopa

The applicant explained istradefylline interactions with levodopa/carbidopa as follows:

In a rat study, the AUC of istradefylline was higher after administration of istradefylline in combination with levodopa/carbidopa compared with istradefylline alone and the increases in the AUC of istradefylline were greater with higher doses of istradefylline. However, since levodopa/carbidopa had no effect on the AUC ratio of M1 to istradefylline or the plasma concentration-time profile after intravenous administration of istradefylline, it was considered that levodopa/carbidopa has no effect on the metabolism or distribution of istradefylline. On the other hand, in a study with ^{14}C -istradefylline, when a high dose of istradefylline was administered in combination with levodopa/carbidopa, the absorption rate was increased. Thus, it was considered that istradefylline interactions with levodopa/carbidopa in rats involve a change in the absorption of istradefylline. As levodopa inhibits gastric emptying, it seems that levodopa slowed down the passage of istradefylline from the stomach to the intestinal tract, resulting in an increased amount absorbed and an increase in the AUC of istradefylline.

PMDA considers as follows:

In light of the applicant's explanation, also in humans, levodopa/carbidopa may increase the absorption rate of istradefylline, resulting in an increase in the plasma concentration of istradefylline. Istradefylline interactions with levodopa/carbidopa in humans will be reviewed in "4.(ii) Summary of clinical pharmacology studies."

3.(iii) Summary of toxicology studies

3.(iii).A Summary of the submitted data

Toxicity studies of istradefylline conducted include single-dose toxicity, repeat-dose toxicity, genotoxicity, carcinogenicity, reproductive and developmental toxicity, and other toxicity (antigenicity, the mechanism of toxicity, dependence, phototoxicity, toxicity of istradefylline in combination with levodopa/carbidopa) studies.

3.(iii).A.(1) Single-dose toxicity (Attached document 4.2.3.1-1 to 4.2.3.1-5, 4.2.3.1-8)

Single-dose toxicity studies were conducted by the oral route of administration in mice, rats, dogs, and monkeys and the approximate lethal doses were determined to be >2000 mg/kg in mice, >2000 mg/kg in rats, >1200 mg/kg in dogs, and >2700 mg/kg in monkeys. As symptoms after administration, increased spontaneous locomotor activity and excitement were observed in mice and rats while there were no apparent toxicological signs in dogs and monkeys.

3.(iii).A.(2) Repeat-dose toxicity

Repeat-dose toxicity studies were conducted by the oral route of administration in rats (4-week and 26-week studies) and dogs (4-week, 26-week, and 52-week studies). The primary target organs of toxicity were the lungs (rats, dogs), adrenal gland (rats, dogs), liver (rats, dogs), brain (rats), pancreatic exocrine portion (rats), kidneys (rats), heart and blood vessels (rats), skeletal muscle (rats), lymphoid and hematopoietic organs (rats, dogs), and exocrine gland (rats) etc. The ratio of the no-observed-effect-level in non-clinical studies to the clinical exposure was <1 for lung effects (accumulation of macrophages/foamy macrophages/histiocytes/foamy histiocytes in the alveolar spaces and pneumonia) and adrenal effects (hypertrophy of the zona glomerulosa of the adrenal cortex; hypertrophy, swelling, and hyperplasia of the zona fasciculata of the adrenal cortex) in both rats and dogs. In rats, mineralization in the brain, i.e. mineralization in the walls of small arteries and capillaries in the thalamus and midbrain were observed. In rats, increased spontaneous locomotor activity, which is considered related to the pharmacological effect of istradefylline, was noted.

The exposure of istradefylline at the no-observed-adverse-effect level (NOAEL) in rats (a 26-week study) (30 mg/kg/day) (males, 11,482 ng·h/mL; females, 6869 ng·h/mL) was 0.9- to 1.4-fold the human clinical exposure (7925 ng·h/mL). The exposure of istradefylline at the NOAEL in dogs (a 52-week study) (10 mg/kg/day) (males, 6556 ng·h/mL; females, 6284 ng·h/mL) was 0.8-fold the human clinical exposure.

3.(iii).A.(2).1 Rat 4-week repeated oral dose toxicity study (1) (Attached document 4.2.3.2-2)

Istradefylline (0 [vehicle], 6, 30, 160, 800 mg/kg/day) was orally administered to male and female SD rats (n = 15/sex/group) for 4 weeks. No death occurred. Reduced body weight gain, decreased food consumption, and increases in blood cholesterol (Cho) and phospholipids (PL) in males at 6 mg/kg/day; increased spontaneous locomotor activity in males and females at ≥6 mg/kg/day (excluding males in the 800 mg/kg/day group); increased food consumption or a trend towards increased food consumption in males and females at

≥30 mg/kg/day; a trend towards increased body weight, an increase in blood alanine aminotransferase (ALT) and a decrease in Cl/Na ratio, and increased liver weight or a trend towards increased liver weight in females at ≥30 mg/kg/day; an increase in urine volume in males at 160 mg/kg/day; irritability in females at 160 mg/kg/day; an increase in blood aspartate aminotransferase (AST) and a decrease in blood potassium, increased adrenal gland weight, and swelling of adrenal zona fasciculata cells in females at ≥160 mg/kg/day; an increase in blood Cho, increased kidney weight, light-colored kidneys, vacuolation of the proximal tubular epithelium, and increased single cell necrosis and vacuolation of acinar cells of the pancreas in males and females at 800 mg/kg/day; increases in blood AST and PL, decreases in α1-globulin fraction and K, an increase in urine pH, increased adrenal gland weight, and swelling of adrenal zona fasciculata cells in males at 800 mg/kg/day; and increases in blood glucose (Glu), total bilirubin (T-Bil), and albumin fraction and a decrease in blood K in females at 800 mg/kg/day were observed. The increased values for clinical chemistry parameters (ALT, AST, T-Bil, Cho, PL) were all less than 2-fold those of the vehicle control group. Although food consumption was decreased in males of the 6 and 30 mg/kg/day groups in the early phase of the recovery period, all changes excluding increased kidney weight resolved during the 4-week recovery period. As the increased values for clinical chemistry parameters (ALT, AST, T-Bil, Cho, PL) were all less than 2-fold those of the vehicle control group, the NOAEL was determined to be 30 mg/kg/day.

3.(iii).A.(2).2 Rat 4-week repeated oral dose toxicity study (2) (Attached document 4.2.3.2-11)

istradefylline (0 [vehicle], 25, 100, 400 mg/kg/day) was orally administered to male and female SD rats (n = 10/sex/group) for 4 weeks. No treatment-related death occurred. Increases in blood T-Bil and Cho, an increase in urine volume, and vacuolation of acinar cells of the pancreas in males and females at ≥100 mg/kg/day; increased lung and kidney weights, increased alveolar macrophages, hypertrophy of the adrenal zona fasciculata, and apoptosis of acinar cells of the pancreas in males at ≥100 mg/kg/day; increases in the reticulocyte rate and urine pH, alopecia, and increased adrenal gland and spleen weights in females at ≥100 mg/kg/day; increases in blood AST and Glu, increased liver weight, microvacuolization of the cortical tubules of the kidney, and pneumonia associated with thickened alveolar septum in males and females at 400 mg/kg/day; an increase in blood PL, decreased fat tissue, increased adrenal gland and heart weights, centrilobular hepatocellular hypertrophy in the liver, decreased prostate gland weight, and decreased colloid content in males at 400 mg/kg/day; and an increase in blood ALT, swelling of the adrenal gland, hypertrophy of the adrenal zona fasciculata, increased alveolar macrophages, and apoptosis of acinar cells of the pancreas in females at 400 mg/kg/day were observed. Electron microscopy of the lungs of rats treated with 400 mg/kg/day of istradefylline revealed vesicles, multilayered cellular debris, or tubular myelin within alveolar macrophages and tubular myelin in the alveolar spaces. Based on the above, the NOAEL was determined to be 25 mg/kg/day.

3.(iii).A.(2).3 Rat 26-week repeated oral dose toxicity study (Attached document 4.2.3.2-5)

Istradefylline (0 [vehicle], 3, 6, 30, 160 mg/kg/day) was orally administered to male and female SD rats (n = 15/sex/group) for 26 weeks. No death occurred. Increased spontaneous locomotor activity and increases in gait, grooming, and rearing etc. in males and females at ≥3 mg/kg/day; increases in body weight and food consumption or a trend towards increases in body weight and food consumption in females at ≥3 mg/kg/day,

irritability in females at ≥ 6 mg/kg/day; a decrease in blood K in males at ≥ 30 mg/kg/day; increases in blood direct bilirubin (D-Bil) and T-Bil or a trend towards increases in blood D-Bil and T-Bil and cerebral mineralization in males and females at 160 mg/kg/day, irritability, increases in body weight and food consumption or a trend towards increases in body weight and food consumption, increased kidney weight, and swelling of the liver and light-colored liver and centrilobular hepatocellular fatty changes in males at 160 mg/kg/day; and a decrease in platelet count, reduced prothrombin time, decreases in blood Cl and Cl/Na ratio, increased adrenal gland weight and swelling of zona fasciculata cells, and increased liver weight in females at 160 mg/kg/day were observed. The mineralized deposits were found in the walls of small arteries and capillaries in the thalamus and midbrain and polysaccharides and calcium salts were detected by periodic acid-Schiff (PAS) staining, Kossa staining, and alizarin red staining and the absence of lesions in the myelin sheath and neurons was confirmed. Based on the above, the NOAEL was determined to be 30 mg/kg/day.

3.(iii).A.(2).4 Dog 4-week repeated oral dose toxicity study (1) (Attached document 4.2.3.2-17)

Istradefylline packed in a gelatin capsule (0 [vehicle], 25, 100, 400 mg/kg/day) or an empty capsule was orally administered to male and female beagle dogs (n = 4/sex/group) for 4 weeks. Increased adrenal gland weight in females at 100 mg/kg/day; a trend towards decreased body weight and increases in blood ALT, AST, D-Bil, and T-Bil or a trend towards increases in blood ALT, AST, D-Bil, and T-Bil in males and females at 400 mg/kg/day; increases in blood Cho and urine protein in males at 400 mg/kg/day; and a trend towards decreased food consumption in females at 400 mg/kg/day were observed. After a 4-week recovery period, all changes resolved. The increased adrenal gland weight observed at 100 mg/kg/day was not associated with histopathological changes and the NOAEL was determined to be 100 mg/kg/day.

3.(iii).A.(2).5 Dog 4-week repeated oral dose toxicity study (2) (Attached document 4.2.3.2-22)

Istradefylline packed in a gelatin capsule (30, 100, 300 mg/kg/day) or an empty capsule was orally administered to male and female beagle dogs (n = 3/sex/group) for 4 weeks. Hypertrophy of the adrenal zona fasciculata in males and females at ≥ 100 mg/kg/day; light-colored stool or watery light-colored stool, decreased body weight gain, and a trend towards increased liver weight in males at ≥ 100 mg/kg/day; a trend towards decreased pancreas weight, increased macrophages in the alveolar spaces, and pneumonia associated with thickened alveolar septum in females at ≥ 100 mg/kg/day; increased blood Cho or a trend towards increased blood Cho, a trend towards increased adrenal gland weight, and centrilobular hepatocellular hypertrophy and clear cytoplasm of hepatocytes in the liver in males and females at 300 mg/kg/day; increases in blood ALT and AST, increased macrophages in the alveolar spaces, and pneumonia associated with thickened alveolar septum in males at 300 mg/kg/day; and light-colored stool or watery light-colored stool, decreased food consumption, and an increase in the density of interstitial cells and atrophy of the exocrine gland of the pancreas in females at 300 mg/kg/day were observed. Based on the above, the NOAEL was determined to be 30 mg/kg/day.

3.(iii).A.(2).6 Dog 26-week repeated oral dose toxicity study (Attached document 4.2.3.2-19)

Istradefylline packed in a gelatin capsule (0 [vehicle], 16, 80, 400 mg/kg/day) or an empty capsule was orally administered to male and female beagle dogs (n = 4/sex/group) for 26 weeks. Swelling of the adrenal gland,

hyperplasia of zona fasciculata cells of the adrenal cortex, and bile thrombi in the bile canaliculi of the liver in females at ≥ 80 mg/kg/day; increases in blood ALT, AST, D-Bil, and T-Bil or a trend towards increases in blood ALT, AST, D-Bil, and T-Bil, increased adrenal gland weight, and macrophage infiltration into the alveolar spaces in males and females at 400 mg/kg/day; swelling of the adrenal gland in males at 400 mg/kg/day; and a trend towards increases in blood ALP and Cho and hepatocellular vacuolation in females at 400 mg/kg/day were observed. Based on the above, the NOAEL was determined to be 16 mg/kg/day.

3.(iii).A.(2).7) Dog 52-week repeated oral dose toxicity study (Attached document 4.2.3.2-26)

Istradefylline packed in a gelatin capsule (10, 30, 100 mg/kg/day) or an empty capsule was orally administered to male and female beagle dogs ($n = 5/\text{sex}/\text{group}$) for 52 weeks. As one male dog in the 30 mg/kg/day group exhibited anastasia, convulsion, and hyperthermia at Week 42, the dog was sacrificed in extremis and the histopathological findings revealed idiopathic meningeal polyarteritis, which was an incidental finding unrelated to istradefylline. Swelling of the adrenal gland in females at ≥ 10 mg/kg/day; increased pituitary gland weight in males at ≥ 30 mg/kg/day; increased adrenal gland weight or a trend towards increased adrenal gland weight and hypertrophy of the adrenal zona fasciculata in females at ≥ 30 mg/kg/day; increased white blood cell count or a trend towards increased white blood cell count, increases in blood ALT, AST, and Cho or a trend towards increases in blood ALT, AST, and Cho, and increased alveolar macrophages in males and females at 100 mg/kg/day; a trend towards increased adrenal gland weight, swelling of the adrenal gland, and hypertrophy of the adrenal zona fasciculata in males at 100 mg/kg/day; and a trend towards decreased body weight gain in females at 100 mg/kg/day were observed. After a 12-week recovery period, the changes in the adrenal gland tended to resolve and other changes resolved. The swelling of the adrenal gland observed in the 10 mg/kg/day group was not associated with histopathological changes and the NOAEL was determined to be 10 mg/kg/day.

3.(iii).A.(3) Genotoxicity (Attached document 4.2.3.3.1-1 to 4.2.3.3.1-3, 4.2.3.3.2-1, 4.2.3.3.2-3)

Istradefylline was negative for genotoxicity in bacterial reverse mutation assays, a chromosomal aberration assay using cultured mammalian cells (Chinese hamster lung fibroblast cell line), an oral mouse bone marrow micronucleus assay, and a rat primary hepatocyte unscheduled DNA synthesis assay.

3.(iii).A.(4) Carcinogenicity

3.(iii).A.(4).1) Carcinogenicity study in mice (Attached document 4.2.3.4.1-7)

Istradefylline (0 [vehicle], 25, 125, 250 mg/kg/day) was orally administered to male and female ICR mice ($n = 69/\text{sex}/\text{group}$) for 104 weeks. There was no treatment-related increase in tumor incidence and it was concluded that istradefylline is not carcinogenic in mice. Although the incidence of vascular tumors tended to increase in females (0 mg/kg/day, 0 of 69 mice; 25 mg/kg/day, 1 of 69 mice; 125 mg/kg/day, 2 of 69 mice; 250 mg/kg/day, 4 of 69 mice), the laboratory background incidence in females (in the 8 years proximate to the study year, 1996-2003; 11 studies, a total of 966 mice) was 4.0% (the incidence per study ranged from 0.0% to 8.8%) and vascular tumor is not a rare tumor in the strain of mice used and the statistical criterion

suggested in the guidelines for common tumors ($P < 0.01^*$ for a pairwise comparison) was not met. Thus, it was concluded that the increases in vascular tumors are not treatment-related. As non-neoplastic changes, pigmentation of the adrenal medulla, pneumonia, vacuolation of acinar cells of the pancreas, histiocytes indicative of pigmented pancreas, and vacuolation of myocytes in the skeletal muscle in males and females at ≥ 125 mg/kg/day; adenosis and mineralization of the lacrimal gland and hepatocellular hypertrophy in males at ≥ 125 mg/kg/day; histiocytes indicative of pigmented liver in females at ≥ 125 mg/kg/day; eosinophilic histiocytes in the lung in males and females at 250 mg/kg/day; fibrosis of the salivary gland in males at 250 mg/kg/day; and hepatocellular hypertrophy and foamy histiocytes in the lung in females at 250 mg/kg/day were observed.

3.(iii).A.(4).2) Carcinogenicity study in rats (Attached document 4.2.3.4.1-11)

Istradefylline (0 [vehicle], 30, 100, 320 mg/kg/day) was orally administered to male and female Wistar rats ($n = 52/\text{sex}/\text{group}$) for 104 weeks. As a result, an increase in the incidence of breast adenocarcinomas was observed in the 320 mg/kg/day group (0 mg/kg/day, 5 of 52 rats; 30 mg/kg/day, 1 of 52 rats; 100 mg/kg/day, 7 of 52 rats; 320 mg/kg/day, 10 of 52 rats). The laboratory background incidence of breast adenocarcinomas (in the 11 years proximate to the study year, 1995-2005; 29 studies; a total of 1574 female rats) was 4.1% (the incidence per study ranged from 0.0% to 8.0%) and the incidences in the istradefylline-treated groups as well as in the control group exceeded the range of the historical data at the laboratory. Although the incidence of thyroidal follicular adenomas tended to increase (0 mg/kg/day, 5 of 52 male rats; 30 mg/kg/day, 5 of 52 male rats, 100 mg/kg/day, 8 of 52 male rats; 320 mg/kg/day, 9 of 52 male rats; 0 mg/kg/day, 1 of 52 female rats, 30 mg/kg/day, 0 of 52 female rats; 100 mg/kg/day, 4 of 52 female rats; 320 mg/kg/day, 4 of 52 female rats), the laboratory background incidences (in the 11 years proximate to the study year, 1995-2005; 29 studies; a total of 1572 male rats; a total of 1570 female rats) were 7.2% in males (the incidence per study ranged from 0.0% to 18.0%) and 3.2% in females (the incidence per study ranged from 0.0% to 11.7%) and thyroidal follicular adenoma is not a rare tumor in the strain of rats used and the statistical criterion suggested in the guidelines for common tumors ($P < 0.01$ for a pairwise comparison) was not met. Thus, it was concluded that the increases in thyroidal follicular adenomas are not treatment-related.

Survival was decreased in istradefylline-treated rats, i.e. 83% in males and 75% in females in the vehicle control group vs. 46% in males and 33% in females in the 320 mg/kg group. The causes of death in the 320 mg/kg group were mainly non-neoplastic lung lesions in both males and females and breast adenocarcinoma and pituitary tumor in females. There was a dose-dependent increase in lung lesions and the incidence of pituitary tumors was not significantly different between the 320 mg/kg group (4 of 52 male rats; 25 of 52 female rats) and the control group (11 of 52 male rats; 24 of 52 female rats) in either males or females.

As non-neoplastic changes, mineralization in the brain and inflammatory changes, mainly alveolar macrophage accumulation in the lung in males and females at ≥ 30 mg/kg/day; myocardial fibrosis in females

* FDA. Guidance for industry, Statistical Aspects of the Design, Analysis, and Interpretation of Chronic Rodent Carcinogenicity Studies of Pharmaceuticals (Draft Guidance). 2001.

at ≥ 30 mg/kg/day; vacuolation of the adrenal cortex in males and females at ≥ 100 mg/kg/day; centrilobular hypertrophy in the liver in males at ≥ 100 mg/kg/day; eosinophilic inclusion body in the olfactory epithelium of the nasal cavity, degeneration and necrosis of the skeletal muscle, chronic progressive nephropathy, and an absence of corpora lutea in the ovary in females at ≥ 100 mg/kg/day; bone marrow hyperplasia, diffuse hyperplasia of the parathyroid gland, arteritis, and periarteritis in males and females at 320 mg/kg/day; degeneration of sperm cells in the epididymal lumina and skeletal muscle myofibrillar degeneration and necrosis in males at 320 mg/kg/day; and aneurysmal dilatation in the pancreas, adenomyosis of the uterus, cystic dilatation of endometrial glands, squamous epithelial cysts of the uterine cervix, apoptosis of acinar cells of the pancreas, and enlarged ovarian cyst in females at 320 mg/kg/day were observed.

3.(iii).A.(5) Reproductive and developmental toxicity

As reproductive and developmental toxicity studies, rat studies of fertility and early embryonic development to implantation, embryo-fetal development studies in rats and rabbits, and rat studies on pre- and postnatal development, including maternal function were conducted. In the study of fertility and early embryonic development to implantation, smaller testis and epididymis, decreased sperm motility, a trend towards decreased implantation rate, and decreased fertility index etc. were observed. In the embryo-fetal development studies, a decreased rate of live fetuses, decreased fetal body weight, skeletal variations (split ossification center in the cervical vertebral arch), and gross external anomalies (microphthalmia and ectrodactyly) etc. were observed. In the study on pre- and postnatal development, including maternal function, a trend towards decreases in postweaning survival rate and pup body weight was observed. The exposure at the NOAEL for embryo-fetal development (200 mg/kg/day in both rats and rabbits) was approximately 3.5-fold (rats) and 3.1-fold (rabbits) the human clinical exposure. Istradefylline has been shown to cross the placenta and be excreted in milk [see “3.(ii).A.(2) Distribution and 3.(ii).A.(4) Excretion”] and it was concluded that the findings in the pups were due to their exposure to istradefylline via the placenta or milk.

3.(iii).A.(5).1 Rat study of fertility and early embryonic development to implantation (Attached document 4.2.3.5.1-2)

Istradefylline (0 [vehicle], 160, 360, 800 mg/kg/day) was orally administered to male and female SD rats (n = 16/sex/group). Males were dosed from 4 weeks prior to mating until 6 weeks after the initial mating and females were dosed from 2 weeks prior to mating until gestation day 7. Male rats treated for 4 weeks were mated with treated female rats and the same male rats treated for about 8 weeks were mated with untreated female rats. Locomotor activity/resting position other than sleep and increased spontaneous locomotor activity, increased body weight or a trend towards increased body weight, increased body weight gain or a trend towards increased body weight gain, and increased food consumption or a trend towards increased food consumption in males and females at ≥ 160 mg/kg/day and increased adrenal gland weight or a trend towards increased adrenal gland weight in males at ≥ 160 mg/kg/day were observed. In male animals, decreased fertility index, swelling of the kidney, smaller testis and epididymis, and decreased sperm motility as detected on a sperm test were observed at 800 mg/kg/day, but embryonic development was unaffected. In female animals, a trend towards decreased implantation rate at ≥ 360 mg/kg/day and decreased fertility index at 800 mg/kg/day were observed, but embryonic development was unaffected. Based on the above, the

NOAELs for paternal and maternal general toxicity were determined to be <160 mg/kg/day, the NOAEL for reproductive toxicity was determined to be 160 mg/kg/day, and the NOAEL for embryonic development was determined to be 800 mg/kg/day.

3.(iii).A.(5).2) Embryo-fetal development study in rats (Attached document 4.2.3.5.2-2)

Istradefylline (0 [vehicle], 40, 200, 1000 mg/kg/day) was orally administered to pregnant SD rats (n = 20/group) from gestation day 7 to gestation day 17. Increases in spontaneous locomotor activity and food consumption at ≥ 40 mg/kg/day, increased body weight gain or a trend towards increased body weight gain throughout the dosing period and decreased body weight gain after the dosing period at 40 and 200 mg/kg/day, decreased food consumption after the dosing period at 40 mg/kg/day, and irritability and a transient decrease in body weight gain in the early phase of dosing at 1000 mg/kg/day were observed. In the fetuses, decreased body weight and an increased incidence of skeletal variations (split ossification center in the cervical vertebral arch) were observed at 1000 mg/kg/day. It has been discussed that skeletal variations disappear after delivery because no abnormalities were detected in pups from the 1000 mg/kg/day group on postnatal days 4 and 22 in a dose-finding study for a rat study on pre- and postnatal development, including maternal function (Attached document 4.2.3.5.3-1 [Reference data]). Based on the above, the NOAEL for maternal general toxicity was determined to be <40 mg/kg/day, the NOAEL for reproductive toxicity was determined to be 1000 mg/kg/day, and the NOAEL for embryo-fetal development was determined to be 200 mg/kg/day.

3.(iii).A.(5).3) Embryo-fetal development study in rabbits (Attached document 4.2.3.5.2-3 to 4.2.3.5.2-4)

Istradefylline (0 [vehicle], 50, 200, 800 mg/kg/day) was orally administered to pregnant JW rabbits (n = 20/group) from gestation day 6 to gestation day 18. One rabbit in the 50 mg/kg/day group was found dead on gestation day 17 and it was considered that the cause of death was a gavage error. A trend towards increased food consumption during the dosing period and a trend towards decreased body weight gain after the dosing period at 50 and 200 mg/kg/day and decreased body weight gain, a trend towards decreased food consumption, and decreased body weight gain after the dosing period at 800 mg/kg/day were observed. A decreased rate of live fetuses, decreases in live fetal body weight and placenta weight, and gross external anomalies (microphthalmia and ectrodactyly) were noted in the 800 mg/kg/day group. In the fetuses, the incidence of skeletal anomalies (fused sternum, abnormal alignment of the thoracic spine, fused coccyx, etc.) tended to increase at ≥ 200 mg/kg/day, but there were no statistically significant differences from the vehicle control group and the incidences of individual skeletal anomalies were largely within the range of the historical data at the laboratory. Thus, it was concluded that this effect was not treatment-related. Based on the above, the NOAEL for maternal general toxicity was determined to be <50 mg/kg/day, the NOAEL for reproductive toxicity was determined to be 800 mg/kg/day, and the NOAEL for embryo-fetal development was determined to be 200 mg/kg/day.

In a dose-finding study for this study, adactyly, ectrodactyly, microgenia, and complex malformation (hydrocephaly, microgenia, microphthalmia) were observed after administration of 800 mg/kg/day of

istradefylline to pregnant rabbits.

3.(iii).A.(5).4 Rat study on pre- and postnatal development, including maternal function (Attached document 4.2.3.5.3-3)

Istradefylline (0 [vehicle], 6, 25, 100, 400 mg/kg/day) was orally administered to pregnant SD rats (n = 20/group) from gestation day 7 to lactation day 21. Increased food consumption at 6 mg/kg/day, a transient increase in body weight at 25 mg/kg/day, an increase in the number of dams with total litter loss during lactation at 100 mg/kg/day, and reduced feces, emaciation, and decreased spontaneous locomotor activity after parturition at 400 mg/kg/day were observed. In the pups during lactation, decreases in body weight and body weight gain or a trend towards decreases in body weight and body weight gain at 6 mg/kg/day, a trend towards decreased survival rate on lactation day 4 at ≥ 25 mg/kg/day, and a trend towards decreases in survival rate on lactation day 22 and body weight at 400 mg/kg/day were observed. Based on the above, the NOAELs were determined to be 100 mg/kg/day for maternal general toxicity and 25 mg/kg/day for maternal reproductive toxicity. The NOAEL for offspring was determined to be 6 mg/kg/day, based on the trend towards decreased survival rate on lactation day 4, as the decreased body weight gain was transient.

3.(iii).A.(6) Other toxicity studies

3.(iii).A.(6).1 Antigenicity

As antigenicity studies of istradefylline, passive cutaneous anaphylaxis (PCA) and active systemic anaphylaxis (ASA) tests were performed and it was concluded that istradefylline is unlikely to be antigenic.

(a) Rat passive cutaneous anaphylaxis test with sera from sensitized mice (Attached document 4.2.3.7.1-1)

Male Balb/c mice (n = 5/group) were sensitized by three weekly intraperitoneal injections of 0.01 and 0.1 mg of istradefylline with aluminum hydroxide gel. One week after the last sensitization, sera were collected and intradermally injected to male SD rats. Twenty-four hours later, 0.2 mL of istradefylline solution (5 mg/mL) and 1% Evans blue solution were injected intravenously. As a result, no PCA reaction in response to istradefylline was observed.

(b) Guinea pig active systemic anaphylaxis test and homologous passive cutaneous anaphylaxis test with sera from sensitized guinea pigs (Attached document 4.2.3.7.1-2)

Male Hartley guinea pigs (n = 5/group) were sensitized by three weekly intramuscular injections of 0.1 and 1 mg of istradefylline with Freund's complete adjuvant and 15 days after the last sensitization, 0.2 mL of istradefylline solution (15 mg/mL) was intravenously injected. As a result, no ASA reaction was observed with istradefylline.

Sera were collected from guinea pigs sensitized for ASA reaction 13 days after the last sensitization and intradermally injected to male Hartley guinea pigs. Four hours later, 0.2 mL of istradefylline solution (15 mg/mL) and 1% Evans blue solution were intravenously injected. As a result, no PCA reaction in response to istradefylline was observed.

3.(iii).A.(6).2) Mechanistic study of toxicity (Attached document 4.2.3.7.3-2)

Male SD rats (n = 8/timepoint/group) were treated with single oral doses of 0 (vehicle), 2.5, 10, 25, and 100 mg/kg of istradefylline or 100 mg/kg/day of istradefylline orally for 7 or 14 days. Following single-dose administration of istradefylline, increases in blood adrenocorticotrophic hormone (ACTH), corticosterone (CORT), and aldosterone levels or a trend towards increases in ACTH, CORT, and aldosterone levels were observed. Following repeat-dose administration of istradefylline, no increases in these hormone levels were noted. After 13 day-administration of istradefylline, administration of corticotropin-releasing factor induced clear increases in ACTH, CORT, and aldosterone levels and pituitary responsiveness to corticotropin-releasing factor was retained. Based on the above, it was considered that istradefylline stimulation of ACTH secretion from the pituitary gland is involved in the observed effects on the adrenal gland.

3.(iii).A.(6).3) Dependence studies

(a) Physical dependence study in rats (Attached document 4.2.3.7.4-3)

Male SD rats (n = 10/group) were fed istradefylline (0.125 mg/g of diet in Weeks 1 and 2→0.25 mg/g of diet in Weeks 3 and 4 or 0.5 mg/g of diet in Weeks 1 and 2→1.0 mg/g of diet in Weeks 3 and 4) or diazepam (2→4→6→8 mg/g of diet at weekly intervals) in their diet for 4 weeks, followed by a 1-week withdrawal period. A withdrawal syndrome such as reductions in food consumption and body weight was observed in the diazepam group and transient slight reductions in food consumption and body weight in the early phase of withdrawal were observed in the istradefylline group. Regarding the reductions in food consumption and body weight observed in the istradefylline group, it was considered that these findings occurred as rebound phenomena in response to increased food consumption during the dosing period as istradefylline increases food consumption in rodents and as these reductions in food consumption and body weight were clearly smaller than those in the diazepam group, these were not considered a withdrawal syndrome. Therefore, it was concluded that istradefylline is unlikely to produce physical dependence.

(b) Intravenous self-administration study of the reinforcing effect of istradefylline in rhesus monkeys (Attached document 4.2.3.7.4-7)

Male rhesus monkeys (n = 4) were allowed to self-administer cocaine (≥4 days) and saline was substituted for cocaine (≥4 days). Then monkeys were allowed to intravenously self-administer each of vehicle, 0.125, 0.25, and 0.5 mg/kg/infusion of istradefylline (in this order) for 4 days. The number of injections per day was limited to 20. As a result, the mean number of self-administrations per day increased when monkeys were free to self-administer cocaine. When istradefylline was available compared with vehicle, the mean number of self-administrations per day was markedly higher in 2 of the 4 animals, demonstrating a reinforcing effect of istradefylline.

3.(iii).A.(6).4) Phototoxicity studies

Phototoxicity studies conducted include a phototoxicity study using 3T3 mouse fibroblast cells, a single oral dose and 7-day repeated oral dose phototoxicity study in hairless rats, and a 3-day repeated oral dose phototoxicity study in pigmented rats.

(a) Phototoxicity study using 3T3 mouse fibroblast cells (Attached document 4.2.3.7.7-1)

3T3 mouse fibroblast cells were treated with 0.69 to 14 µg/mL of istradefylline and exposed to UVA (8 J/cm²). As a result, UVA exposure did not affect cell viability.

(b) Single oral dose and 7-day repeated oral dose phototoxicity study in hairless rats (Attached document 4.2.3.7.7-2 [Reference data])

Female hairless rats (RORO-n) (n = 6/group) were treated with single oral doses of 0 (vehicle) and 400 mg/kg/day of istradefylline or 0 (vehicle) and 400 mg/kg/day of istradefylline orally for 7 days and the back skin of each rat was exposed to UVA (5-35 J/cm²) at 4 hours after the last dose. As a result, rats treated with a single dose of istradefylline developed mild skin erythema at 24 hours after UVA exposure at ≥30 J/cm² and rats treated with repeated doses of istradefylline developed mild skin erythema at 4 hours after UVA exposure at ≥20 J/cm², suggesting that istradefylline may cause mild phototoxicity.

(c) Three-day repeated oral dose phototoxicity study in pigmented rats (Attached document 4.2.3.7.7-3)

When male Long-Evans rats (n = 5/group) were treated with 0 (vehicle), 25, 50, 100, and 400 mg/kg/day of istradefylline orally for 3 days and the back skin and eyes of each rat were exposed to UV light (half the minimum level of UV exposure required to induce erythema [0.01 J/cm²]) at 6 hours after administration on Day 3, no reactions indicative of phototoxicity were seen at any dose level.

3.(iii).A.(6.5) Toxicity studies of istradefylline in combination with levodopa/carbidopa

In order to assess the toxicity of the combination of istradefylline and levodopa/carbidopa, single oral dose toxicokinetic and 4-week oral toxicity studies in rats, a 13-week oral toxicity study in dogs, and an embryo-fetal development study in rabbits were conducted. As a result, in rats, istradefylline plasma concentrations were higher and toxicity findings occurred at a lower dose level with the combination than with istradefylline alone. No new toxicities emerged with the combination. In the embryo-fetal development study in rabbits, a decreased rate of live fetuses, decreased fetal body weight, and teratogenic effects such as skeletal anomalies, gross external anomalies, and visceral variations were noted with the combination.

(a) Single oral dose toxicokinetic study of istradefylline in combination with levodopa/carbidopa in rats (Attached document 4.2.3.7.7-4)

Male SD rats (n = 9/group) were orally treated with 100 mg/kg/day of istradefylline in combination with 0/0, 250/25, and 250/62.5 mg/kg/day of levodopa/carbidopa. The t_{max} values of istradefylline were 2, 8, and 8 hours, respectively, and the systemic exposures (AUCs) were 17,223 ng·h/mL, 60,763 ng·h/mL, and 49,949 ng·h/mL, respectively, demonstrating that levodopa/carbidopa increased the t_{max} and systemic exposure of istradefylline.

(b) Four-week oral toxicity study of istradefylline in combination with levodopa/carbidopa in rats (1) (Attached document 4.2.3.7.7-5)

Male and female SD rats (n = 10/sex/group) were orally treated with 0, 25, 100, and 400 mg/kg/day of istradefylline in combination with 250/25 mg/kg/day of levodopa/carbidopa for 4 weeks. In the 400/250/25 mg/kg/day group, 9 of 20 rats (4 of 10 males, 5 of 10 females) died by Day 7 and all of the remaining rats were sacrificed in extremis on Day 9. Lung lesions were seen in decedent animals and it was demonstrated that levodopa/carbidopa increased the systemic exposure of istradefylline (400/250/25 mg/kg/day group, 203,867-220,301 ng·h/mL) when compared with istradefylline alone (400 mg/kg/day group, 66,219-75,761 ng·h/mL). It seemed that following administration of the combination of istradefylline and levodopa/carbidopa, the exposure of istradefylline reached a lethal level, leading to death.

In all test substance-treated groups including the levodopa/carbidopa alone group, brown staining, piloerection, hypotonia and salivation, reduced activated thromboplastin time or a trend towards reduced activated thromboplastin time, and a decrease in blood triglycerides or a trend towards a decrease in blood triglycerides in males and females, a trend towards decreased body weight, decreased body weight gain, and decreased urine volume in males, and increased physical activity, aggressive behavior, increased liver weight, increased ovary weight, and increased uterine weight or a trend towards increased uterine weight in females were observed and these changes were considered related to levodopa/carbidopa.

In rats treated with ≥ 25 mg/kg/day of istradefylline in combination with levodopa/carbidopa, an increase in blood total bilirubin, increased kidney weight or a trend towards increased kidney weight, and alveolar macrophage accumulation associated with alveolitis in males and females, decreased white blood cells and basophilic change of the cortical tubules of the kidney in males, and a decrease in hemoglobin and increased adrenal gland weight in females were observed. In rats treated with 100 mg/kg/day of istradefylline in combination with levodopa/carbidopa, increased lung weight or a trend towards increased lung weight and apoptosis of the thymic cortex in males and females, a decrease in mean corpuscular hemoglobin concentration and acinar cell atrophy of the mammary gland in males, and apoptosis and vacuolation of acinar cells of the pancreas and apoptosis in the medulla of submandibular lymph nodes in females were observed. The above changes observed in rats treated with istradefylline in combination with levodopa/carbidopa were due to an increase in the systemic exposure of istradefylline, secondary to deterioration in the general condition, or related to levodopa/carbidopa and no new toxicities emerged with the combination.

(c) Four-week oral toxicity study of istradefylline in combination with levodopa/carbidopa in rats (2) (Attached document 4.2.3.7.7-7)

Male and female SD rats (n = 10/sex/group) were orally treated with 0/0/0, 0/10/1, 0/50/5, 0/250/25, 100/10/1, 100/50/5, 100/250/25, and 100/0/0 mg/kg/day of istradefylline in combination with levodopa/carbidopa for 4 weeks. In the 100/250/25 mg/kg/day group, 4 of 20 rats (0 of 10 males, 4 of 10 females) died and it was demonstrated that levodopa/carbidopa increased the systemic exposure of istradefylline (100/250/25 mg/kg/day group, females, 173,649 ng·h/mL) when compared with istradefylline alone (100 mg/kg/day group, females, 51,015 ng·h/mL). No new toxicities emerged with the combination.

(d) Thirteen-week oral toxicity study of istradefylline in combination with levodopa/carbidopa in dogs (Attached document 4.2.3.7.7-8)

Male and female beagle dogs (n = 4/sex/group) were orally treated with 0, 30, 100, and 300 mg/kg/day of istradefylline packed in a gelatin capsule in combination with 80/20 mg/kg/day of levodopa/carbidopa for 13 weeks. In order to prevent vomiting of istradefylline due to the emetic effects of levodopa/carbidopa, levodopa/carbidopa were given 4 hours after the administration of istradefylline. As a result, one male dog in the levodopa/carbidopa alone group died. In the levodopa/carbidopa alone group, vomiting and salivation and a decrease in blood ALT in males and females, increased food consumption in males, and increased submandibular gland weight or a trend towards increased submandibular gland weight in females were observed. In dogs treated with istradefylline in combination with levodopa/carbidopa, vomiting and salivation and a decrease in blood ALT in males and females, decreased body weight gain, acinar cell hypertrophy of the submandibular gland, and decreased heart rate in males, and increased adrenal gland weight or a trend towards increased adrenal gland weight and increased submandibular gland weight or a trend towards increased submandibular gland weight in females were observed. The decreased heart rate was considered related to levodopa/carbidopa as it occurred 2 hours after the administration of levodopa/carbidopa. The decreased body weight gain observed in males at 100/80/20 mg/kg/day and the increased adrenal gland weight or the trend towards increased adrenal gland weight observed in females at $\geq 100/80/20$ mg/kg/day were not noted after a 4-week recovery period. Levodopa/carbidopa did not affect the systemic exposure of istradefylline when compared with istradefylline alone. Based on the above, no new toxicities emerged with the combination.

(e) Embryo-fetal development study of istradefylline in combination with levodopa/carbidopa in rabbits (Attached document 4.2.3.7.7-12)

Pregnant JW rabbits (n = 20/group) were treated with repeated oral doses of 400/0/0, 50/80/20, 200/80/20, 400/80/20, and 0/80/20 mg/kg/day of istradefylline in combination with levodopa/carbidopa or vehicle. There were no effects on maternal general condition and reproduction and fetal development in the levodopa/carbidopa alone group and a transient decrease in food consumption early after the dosing period in maternal animals and decreased fetal body weight were observed in the istradefylline alone group. In maternal animals treated with istradefylline in combination with levodopa/carbidopa, decreased body weight gain after the dosing period and a transient decrease in food consumption early after the dosing period were observed and in the fetuses, decreased body weight and digit malformations (ectrodactyly, absence of nails, deficiency of phalanges) at $\geq 200/80/20$ mg/kg/day and a decreased rate of live fetuses, decreased placenta weight, and increased incidences of gross external anomalies (adactyly, brachydactyly, etc.), visceral anomalies (deficiency of the membranous ventricular septum, persistent truncus arteriosus), and skeletal anomalies (smaller phalanges) at 400/80/20 mg/kg/day were observed, which were considered treatment-related. Also at 50/80/20 mg/kg/day, one fetus had complex malformation (ectrodactyly, absence of nails, hemimelia, complex anomaly of the skull, deficiency of phalanges), which was considered incidental because complex malformation was not noted with higher doses of istradefylline in combination with levodopa/carbidopa. Levodopa/carbidopa slightly increased the systemic exposure of istradefylline on gestation day 18. Based on the above, no new toxicities emerged with the combination.

3.(iii).B Outline of the review by PMDA

3.(iii).B.(1) Findings in the lungs

PMDA asked the applicant to explain the mechanism of lung lesions observed in toxicity studies of istradefylline.

The applicant responded as follows:

Since it is known that the adenosine A_{2A} receptor, which is the target for istradefylline, is distributed in the lungs (Dixon AK et al., *Br J Pharmacol.* 1996;118:1461-8, Yan L et al., *Exp Opin Emerging Drug.* 2003;8:537-76), though the mechanism is not defined, istradefylline itself may affect the lungs. In repeat-dose toxicity studies in rats and dogs, electron microscopy revealed myelin or multilayered structures within alveolar macrophages or type II alveolar epithelial cells. Thus, lung lesions may have been caused by accumulation of surfactants due to overproduction of surfactants by type II alveolar epithelial cells or alveolar macrophage dysfunction. In rats, proliferation of type II alveolar epithelial cells was observed, which is considered to occur following a loss of type I alveolar epithelial cells. Thus, istradefylline may have affected type I alveolar epithelial cells. Furthermore, possible causes not directly related to istradefylline include a response to a foreign body due to the aspiration of the dosing solution or istradefylline packed in a gelatin capsule or vehicle because the observed lung effects included foamy macrophages.

PMDA asked the applicant to discuss the safety of istradefylline in the lungs of humans and explain the need to include a caution statement regarding the lung effects observed in toxicity studies in the package insert and take post-marketing measures.

The applicant responded as follows:

If inflammatory changes in the lungs, mainly macrophages, as observed in toxicity studies, persist for a long period of time in humans, diseases like alveolar proteinosis or diffuse alveolar damage, as chronic alveolar damage, can develop. Although chronic alveolar damage has not been reported as an adverse event in Japanese clinical studies, testing capable of detecting a change in the lungs was not required in clinical studies and in light of the results of nonclinical studies of istradefylline and the putative mechanisms of lung lesions, the possibility of the occurrence of lung toxicity-related adverse events in humans can not be ruled out. Based on the above, it will be stated in the package insert that inflammatory changes in the lungs were observed in non-clinical studies and furthermore, the package insert will advise that patients should be closely monitored after initiating treatment with istradefylline and if shortness of breath/dyspnoea or dry cough occurs, imaging studies such as chest X-ray or an appropriate work-up etc. should be performed and the dose should be reduced, the drug should be interrupted or discontinued, or other appropriate therapeutic measures should be taken, as needed. After the market launch, efforts will be made to identify the causes for shortness of breath, dyspnoea, dry cough, and pneumonia reported by patients treated with istradefylline.

PMDA considers as follows:

The adenosine A_{2A} receptor, which is the target for istradefylline, is distributed also in tissues other than the

lungs and the direct mechanism of lung lesions is unknown. Lung lesions developed in all animal species tested for lung effects of istradefylline, the exposure at the NOAEL for increased pneumonia in a dog 4-week repeated oral dose toxicity study was 1.0- to 1.1-fold the human clinical exposure and the NOAELs for lung effects identified in some other toxicity studies also yielded safety margins of <3 , and when istradefylline was administered for 104 weeks in a rat carcinogenicity study, lung effects occurred at a lower dose level than in the above dog study, causing deterioration of the general condition and death. Therefore, there is no adequate safety margin for lung effects. In addition, testing capable of detecting a change in the lungs was not required in clinical studies and in view of the limited ability of clinical studies to detect lung effects, the possibility that adverse events related to lung toxicity observed in toxicity studies occur in humans can not be ruled out. Clinical studies conducted with istradefylline at doses of 20 and 40 mg have suggested no problem considered related to chronic alveolar damage [see “4.(iii) Summary of clinical efficacy and safety”]. In foreign clinical studies of ≥ 52 -week duration, cough, dyspnoea, and pneumonia etc. have been reported as adverse events, but the progression of lesions like chronic alveolar damage has not been observed. Taking account of these findings etc., istradefylline can be indicated for clinical use, but it is necessary to provide adequate caution in the package insert. The precaution statements proposed by the applicant will be finalized, taking also account of comments from the Expert Discussion.

3.(iii).B.(2) Findings in the brain

PMDA asked the applicant to explain the mechanism of mineralization in the walls of small arteries and capillaries in the thalamus and midbrain of rats and the clinical risk of this finding.

The applicant responded as follows:

An increased incidence and severity of mineralization in the brain with istradefylline was observed in the rat only among the animal species used in toxicity studies. Although the origin of the mineralized deposits in the brain and the mechanism of the species differences are unknown, these mineralized deposits in the brain were morphologically similar to age-related ones. Although the main components of these mineralized deposits in the brain were calcium and phosphorus, there were no consistent changes in blood calcium or inorganic phosphate and systemic involvement was absent. Therefore, this was unlikely to be metastatic mineralization and was considered possibly related to local formation of blood vessels or metabolism at favorite sites in the brain. As the finding was not associated with changes in the parenchyma of nervous tissue (neuronal necrosis, degeneration, and inflammatory changes, glial appearance, hemorrhage, etc.) and was asymptomatic and occurred in the rat only, this is unlikely to be clinically relevant. Also in humans, vascular mineralization, primarily in the basal ganglia, is observed in healthy elderly persons and patients with central nervous system diseases including Parkinson's disease or diseases with thyroid dysfunction, but is an asymptomatic event.

Since no test method can precisely assess the development and progression of mineralization in the brain at present and it is difficult to identify clinical symptoms related to mineralization in the brain, efforts will be made to collect information on this finding via literature search etc. and if new information becomes available in future, the pharmacovigilance plan for istradefylline will be reviewed. This finding will be noted in the package insert for istradefylline.

PMDA considered as follows and accepted the response:

As the mechanism of the species differences in mineralization in the brain is unknown and the exposure at the NOAEL for this finding was less than 3.3-fold the human clinical exposure, the possibility that similar changes occur also in humans can not be ruled out, but since rats with mineralization in the brain were asymptomatic, there should be no major concern from a toxicological point of view at present. However, no test method can precisely assess the development and progression of mineralization in the brain at present and the clinical significance of mineralization in the brain is not fully understood. Thus, providing caution about the observed toxicity through the package insert etc. can contribute to the management of the risk of this finding.

3.(iii).B.(3) Findings in the adrenal cortex

In repeat-dose toxicity studies in rats and dogs, changes indicative of increased steroid hormone secretion, e.g. increased adrenal gland weight and hypertrophy of zona fasciculata cells were observed and the exposure at the NOAEL for effects on the adrenal gland was lower than the human clinical exposure. PMDA asked the applicant to explain the mechanism of adrenal toxicity and the risk in humans.

The applicant responded as follows:

Concerning the mechanism of adrenal toxicity, adrenal changes were considered attributable to the effect of istradefylline on the pituitary-adrenocortical axis because hyperactivity of their pituitary-adrenocortical axis in adenosine A_{2A} receptor knockout mice has been reported (Jegou S, et al., *J Neuroendocrinol.* 2003;15:1171-7) and increases in blood ACTH, CORT, and aldosterone levels following a single dose of istradefylline were observed in a mechanistic study of the effect of istradefylline on ACTH and adrenocorticosteroids. Thus, in order to assess the effect of istradefylline on the adrenal gland, blood ACTH and cortisol were measured in a Japanese phase I study. As a result, there were no apparent abnormal changes in ACTH or cortisol levels. Also in Japanese and foreign clinical studies conducted to date, neither adrenal adverse events nor reproductive adverse drug reactions considered related to changes in steroid hormones were reported. Based on the above, hypertrophic changes in the adrenal gland as observed in non-clinical studies are unlikely to occur in humans and are not considered clinically relevant.

PMDA concluded that although the direct mechanism of the adrenal toxicity of istradefylline observed in toxicity studies is not clear, the findings are not clinically relevant and accepted the applicant's response.

3.(iii).B.(4) Carcinogenicity

In a rat carcinogenicity study, the incidence of breast adenocarcinomas tended to increase dose-dependently in rats treated with istradefylline and the incidences in the istradefylline-treated groups exceeded the range of the historical data at the laboratory. PMDA asked the applicant to explain the relationship between istradefylline and breast adenocarcinoma.

The applicant responded as follows:

Based on the data from a rat carcinogenicity study, each istradefylline group was compared with the control group with respect to the time to onset, number, size, and incidence of breast adenocarcinomas, to determine whether istradefylline increases the risk of breast adenocarcinoma. As a result, when the time to observation of the first tumor was compared, the first tumor was observed slightly earlier in a rat of the 320 mg/kg/day group (Week 59) compared with the earliest tumor in the control group (Week 74), which was not considered a biologically meaningful difference because spontaneous tumors developed even earlier (Week 31) at the laboratory. There were no apparent differences in the number or size of adenocarcinomas between the groups. As for comparison of the incidence, the statistical criterion suggested in the guidelines for common tumors ($P < 0.01$ for a pairwise comparison) was not met and istradefylline is not considered associated with an increased risk of breast adenocarcinoma. Based on the above results, compared with the control group, istradefylline is not considered to increase the risk of breast adenocarcinoma in terms of severity and incidence and rat breast adenocarcinomas as well as benign tumors are common tumors also at the laboratory. Therefore, istradefylline is unlikely to induce breast adenocarcinoma.

PMDA's view on the above response is as follows:

The severity and incidence of breast adenocarcinomas observed in a rat carcinogenicity study were not substantially different from those of spontaneous tumors, there was no increase in the number of mice with breast adenocarcinomas in a mouse carcinogenicity study, istradefylline was tested negative for genotoxicity, the incidence of pituitary tumors also was not altered in rats and breast adenocarcinomas were unlikely to be induced by changes in hormone homeostasis, and any mechanism in which istradefylline might directly be involved in increased breast adenocarcinomas in rats has not been identified. Taking account of the totality of these findings etc., istradefylline-related breast adenocarcinoma is unlikely to develop during clinical use.

3.(iii).B.(5) Reproductive and developmental toxicity

The section of "Use during pregnancy, delivery or lactation" in the proposed draft package insert reads that "Administration of the drug to pregnant women or women who may possibly be pregnant is not recommended." In reproductive and developmental toxicity studies of istradefylline alone (rats and rabbits) or in combination with levodopa/carbidopa (rabbits), fetal effects, e.g. teratogenic effects were observed and when the exposure at the NOAEL for embryo-fetal development was compared with the human clinical exposure, safety margins were not adequate, i.e. the rat-to-human safety margin of istradefylline alone was about 3.5-fold, the rabbit-to-human safety margin of istradefylline alone was about 3.1-fold, and the rabbit-to-human safety margin of istradefylline in combination with levodopa/carbidopa was about 1.5-fold. PMDA instructed the applicant to "contraindicate" istradefylline in pregnant women and women who may possibly be pregnant and include specific information on the findings observed in embryo-fetal development studies in the package insert and the applicant responded appropriately.

PMDA considers as follows:

In addition to the above actions, skeletal variations in rats and visceral anomalies suggestive of fetal developmental delay observed in a toxicity study of istradefylline in combination with levodopa/carbidopa, etc., should be mentioned in the package insert. Furthermore, although the incidence of skeletal anomalies tended to increase in rabbits and the applicant concluded that this effect was not a treatment-related toxicity, the possibility that this effect was treatment-related can not be ruled out, comparison with the control group assuming that an effect is related to doses (trend test) showed a significant difference, the incidences in some of the istradefylline-treated groups exceeded the range of the historical data at the laboratory, and a similar toxicological finding, i.e. skeletal variations were determined to be treatment-related also in rats. Therefore, this finding should also be mentioned in the package insert.

3.(iii).B.(6) Dependence potential

Regarding the dependence potential of istradefylline, [REDACTED] was submitted at filing, but [REDACTED]
[REDACTED]
[REDACTED], the applicant [REDACTED]
[REDACTED] not conducted.

Based on “Guidance on Nonclinical Safety Studies for the Conduct of Human Clinical Trials and Marketing Authorization for Pharmaceuticals” (PFSB/ELD Notification No. 0219-4 dated February 19, 2010) and “Scope of Application and Guidelines for Animal Studies and Clinical Observations on Drug Dependence” (Notification No. 113 of the Director of Narcotics Division, Pharmaceutical Affairs Bureau, MHW dated March 14, 1975), PMDA requested the conduct of [REDACTED] and the applicant submitted the results of the evaluation of psychological dependence in rhesus monkeys (Attached document 4.2.3.7.4-7).

PMDA asked the applicant to explain the mechanism of the reinforcing effect of intravenously self-administered istradefylline in rhesus monkeys.

The applicant responded as follows:

The ventral striatum, which is part of the mesolimbic system, extending from the ventral tegmental area to the nucleus accumbens, is known as the reward center of the brain and is known to be involved in the addictive properties of many drugs of abuse. The adenosine A_{2A} receptor is expressed in the ventral striatum (the nucleus accumbens and the olfactory tubercle) and adenosine A_{2A} receptor antagonism by istradefylline in these regions may have been involved in the reinforcing effect of istradefylline. However, it has been reported that the reinforcing effects of drugs such as cocaine, amphetamine, and an opioid are diminished in adenosine A_{2A} receptor knockout mice (Soria G. et al. *Neuropsychopharmacology*. 2006;31: 978-87, Chen JF et al. *Neuropsychopharmacology*. 2003;28:1086-95, Brown RM et al. *Neuropsychopharmacology*. 2009;34:844-56) and the extent to which adenosine A_{2A} receptor antagonism by istradefylline is directly involved in the mechanism of the reinforcing effect of istradefylline is unknown. Istradefylline shows no affinity for the receptors (dopamine, opioid, and cannabinoid receptors) or transporters (monoamine transporters) that are considered involved in drug dependence and has no effect on neurotransmitter metabolizing enzymes (monoamine oxidase, catechol-*O*-methyltransferase) and direct involvement of these receptors or transporters is unlikely.

For the assessment of the similarity to a known dependence-producing drug, a dependence-producing drug with a similar central nervous system effect as the test substance is generally used. Although istradefylline is an analog of caffeine, a xanthine-based compound with a central nervous system stimulating effect, people take caffeine because they prefer it and it is known that users may develop a mild psychological dependence on caffeine and a drug discrimination study of istradefylline was not performed, considering that the information obtained from the study would not be of high toxicological significance.

PMDA considers as follows:

Based on the results from a dependence study, the mechanism of the reinforcing effect of istradefylline (including the degree of the reinforcing effect and generalization) is not clear and abuse potential such as dependence potential has not been evaluated in Japanese and foreign clinical studies. Thus, the possibility that istradefylline intake leads to psychological dependence in humans can not be ruled out and it is necessary to include a precaution statement regarding psychological dependence in the package insert and risk

management in clinical practice is required. It is also necessary to continue to investigate effects in humans via post-marketing surveillance etc. The precaution statement in the package insert and the information to be collected via post-marketing surveillance will be finalized, taking also account of comments from the Expert Discussion.

3.(iii).B.(7) Phototoxicity

In a repeat-dose phototoxicity study in hairless rats, mild skin erythema was induced by ≥ 20 J/cm² of UVA and istradefylline was phototoxic. PMDA asked the applicant to explain safety in humans.

The applicant responded as follows:

A phototoxic dose of 20 J/cm² in this study is comparable to the exposure to sunlight during longer outdoor activities and as the dose level of istradefylline used in this study was 400 mg/kg/day only, the NOAEL for phototoxicity in rats after an excessive dose of UVA irradiation could not be assessed accurately. However, as the skin responses induced by excessive doses of UVA irradiation were mild in severity and the estimated exposure at 400 mg/kg/day in rats was ≥ 10 -fold the systemic exposure following repeated administration of the maximum recommended human dose of istradefylline, the risk of serious phototoxicity in humans should be low. In Japanese clinical studies of istradefylline, as possible phototoxic adverse events, actinic keratosis (1 subject) and erythema (1 subject) occurred, but their causal relationship to istradefylline was denied and there were no strongly suspected cases of phototoxic adverse reactions to istradefylline.

PMDA considered as follows:

Since a 20 J/cm² UVA dose is comparable to the light exposure that humans might be exposed to and the NOAEL for phototoxicity after 20 J/cm² irradiation has not been assessed and istradefylline could induce a phototoxic response even at the clinical dose, the risk of istradefylline-induced phototoxicity in humans can not be excluded.

PMDA instructed the applicant to include this information in the package insert.

The applicant responded that this finding will be mentioned in the package insert and PMDA accepted the applicant's response.

4. Clinical data

4.(i) Summary of biopharmaceutic studies and associated analytical methods

r Summary of the submitted data

In Japanese and foreign clinical studies, plasma concentrations of istradefylline and its metabolites were determined by a validated high performance liquid chromatography-ultraviolet detection (HPLC-UV) method or a validated liquid chromatography/tandem mass spectrometry (LC/MS/MS) method and urine concentrations were determined by a validated HPLC-UV method. The lower limit of quantitation of the HPLC-UV method was 5 ng/mL for both istradefylline and M1 (4'-O-demethyl istradefylline) in plasma, 25 ng/mL for *cis*-istradefylline in plasma, 1 ng/mL for both istradefylline and M1 in urine, and 5 ng/mL for *cis*-

istradefylline in urine. The lower limit of quantitation of the LC/MS/MS method for istradefylline and M1 in plasma differed from study to study, i.e. 0.5 to 1 ng/mL and 0.2 to 1 ng/mL, respectively.

The 20 mg tablets used in a Japanese phase III study are of the same formulation as proposed for marketing. In the Japanese phase III study, subjects in the 20 mg group received two 10-mg tablets of istradefylline and subjects in the 40 mg group received two 20-mg tablets of istradefylline.

Unless otherwise specified, pharmacokinetic parameters are expressed as the mean or the mean \pm SD.

4.(i).A.(1) Bioequivalence study for different dosage form strengths (Study 6002-012, Attached document 5.3.1.2-1)

A two-way crossover study comparing two 10-mg tablets of istradefylline to one 20-mg tablet of istradefylline, which were used in a Japanese phase III study, was conducted in 30 Japanese healthy adult male subjects. Following single oral dose administration under fasted condition (a \geq 12-day washout period), plasma concentrations were measured up to 168 hours post-dose. The geometric mean ratios (two 10-mg tablets of istradefylline/one 20-mg tablet of istradefylline) of C_{\max} and AUC_{0-t} [90% confidence interval (CI)] were 1.068 [1.001-1.140] and 0.985 [0.928-1.046], respectively.

4.(i).A.(2) Food effect

1) Food effect study with the 20-mg tablet (Study 6002-011, Attached document 5.3.1.1-1)

A two-way crossover study using 20 mg of istradefylline in the final to-be-marketed dosage form (the 20-mg tablet) was conducted in 20 Japanese healthy adult male subjects. Following single oral dose administration under fasted and fed conditions (a \geq 21-day washout period), plasma concentrations were measured up to 168 hours post-dose. The median times to reach the maximum plasma concentration of istradefylline (t_{\max}) were 2.00 and 3.00 hours, respectively, the maximum plasma concentrations (C_{\max}) were 112.9 ± 24.1 and 136.4 ± 36.0 ng/mL, respectively, the areas under the plasma concentration-time curve from zero to the last sampling point (AUC_{0-t}) were 3397 ± 1373 and 3833 ± 1465 ng·h/mL, respectively, and the plasma elimination half-lives ($t_{1/2}$) were 57.09 ± 31.51 and 53.56 ± 22.33 hours, respectively, and the geometric mean ratios (fed/fasted) of C_{\max} and AUC_{0-t} [90% CI] were 1.193 [1.115-1.277] and 1.136 [1.055-1.223], respectively.

2) Food effect study with the 50-mg tablet (Study 6002-9601, Attached document 5.3.3.1-1)

A two-way crossover study with the istradefylline 50-mg tablet was conducted in 12 Japanese healthy adult male subjects. Following single oral dose administration under fasted and fed conditions (a 2-week washout period), the median t_{\max} values for istradefylline were 4.00 and 2.00 hours, respectively, the C_{\max} values were 225.5 ± 55.2 and 313.7 ± 42.3 ng/mL, respectively, and the AUC_{0-t} values were 5021 ± 1599 and 5730 ± 1567 ng·h/mL, respectively.

4.(i).B Outline of the review by PMDA

4.(i).B.(1) Bioequivalence of different dosage form strengths

The applicant explained that as the 10-mg tablet and the 20-mg tablet used in a Japanese phase III study exhibited similar dissolution profiles, the efficacy and safety of the istradefylline 20-mg tablet formulation proposed for marketing can be extrapolated from the data from the 20 mg group (two 10-mg tablets) in the Japanese phase III study. However, based on the level of changes in formulation and “Guideline for Bioequivalence Studies for Different Strengths of Oral Solid Dosage Forms” (“BE guideline for different dosage form strengths”) (PFSB/ELD Notification No. 0229-10 dated February 29, 2012), it is difficult to explain the bioequivalence (BE) between the 10-mg tablet and the 20-mg tablet used in the Japanese phase III study by dissolution testing only. Considering that as a rule, it is necessary to determine the BE between the 10-mg tablet and the 20-mg tablet proposed for marketing, which were used in the confirmatory study, in accordance with the BE guideline for different dosage form strengths, PMDA instructed the applicant to explain the BE between the 20-mg tablet proposed for marketing and the 10-mg tablet used in the Japanese phase III study by conducting a human BE study in accordance with “Guideline for Bioequivalence Studies of Generic Products” (“BE guideline for generic products”) (PFSB/ELD Notification No. 0229-10 dated February 29, 2012).

In response to PMDA’s instruction, the applicant conducted a human BE study in accordance with the BE guideline for generic products and explained that BE was demonstrated for the 10-mg tablet and the 20-mg tablet proposed for marketing used in the phase III study.

Based on the study data, PMDA accepted the applicant’s explanation.

4.(ii) Summary of clinical pharmacology studies

4.(ii).A *Summary of the submitted data*

4.(ii).A.(1) Studies using human biomaterials

4.(ii).A.(1).1 Membrane permeability and inhibitory effect on P-glycoprotein (Attached document 4.2.2.2-7 to 4.2.2.2-8)

¹⁴C-istradefylline (0.5 µg/mL), ³H-mannitol, nadolol (100 µg/mL), furosemide (100 µg/mL), propranolol (100 µg/mL), or naproxen (100 µg/mL) (final concentration) was added to the apical (AP) side of Caco-2 cells seeded on transwell insert and incubated for 30 to 120 minutes at 37°C to study apical to basolateral (BL) transport. The apparent permeability coefficient (P_{app}) of istradefylline in the AP-to-BL direction was 32.3×10^{-6} cm/s and the P_{app} values for highly permeable propranolol and naproxen were 22.0×10^{-6} and 107×10^{-6} cm/s, respectively. The ratio of BL-AP P_{app} to AP-BL P_{app} (efflux ratio) of istradefylline was 0.495, less than 1, which was not reduced (0.853) in the presence of quinidine, a P-glycoprotein inhibitor (30 µg/mL). Thus, it was considered that istradefylline is not a substrate for P-glycoprotein.

In the Caco-2 permeability assay, istradefylline inhibited the efflux of digoxin, a substrate for P-glycoprotein, in a concentration-dependent fashion and its 50% inhibitory concentration (IC_{50}) was 0.667 µg/mL.

4.(ii).A.(1).2 Protein binding (Attached document 4.2.2.3-5)

When ³H-istradefylline was added to human serum at final concentrations of 10, 100, and 1000 ng/mL, the

percent protein binding values were 95.0% to 96.6%.

When ^3H -istradefylline at a final concentration of 100 ng/mL was added to 40 mg/mL human serum albumin (HSA) solution, 1 mg/mL human α_1 -acid glycoprotein solution, and 10 mg/mL γ -globulin solution, the percent protein binding values were 94.7%, 66.9%, and 21.4%, respectively.

4.(ii).A.(1).3) Distribution in blood cells (Attached document 5.3.3.1-11 [Reference data])

Following a single oral dose of ^{14}C -istradefylline in 6 foreign healthy adult male subjects, the blood-to-plasma ratio of total radioactivity was 0.57 ± 0.16 to 0.74 ± 0.03 up to 168 hours post-dose.

4.(ii).A.(1).4) *In vitro* metabolism

(a) Metabolism of istradefylline (Attached document 4.2.2.4-8 to 4.2.2.4-10)

Human liver microsomes were incubated with 1 $\mu\text{mol/L}$ of istradefylline in the presence of inhibitors of cytochrome P450 (CYP) isoforms at 37°C for 60 minutes to investigate the effects of CYP inhibitors on the metabolism of istradefylline. In the presence of SKF-525A/1-aminobenzotriazole (nonspecific CYP inhibitors, 100/100 $\mu\text{mol/L}$), ketoconazole (a CYP3A4/5 inhibitor, 1 $\mu\text{mol/L}$), and diethyldithiocarbamate (a CYP2E1 inhibitor, 100 $\mu\text{mol/L}$), the metabolism of istradefylline was inhibited and istradefylline metabolic activities were 23.0%, 26.3%, and 74.3%, respectively, of those in the absence of the CYP inhibitors. Furafylline (a CYP1A2 inhibitor, 10 $\mu\text{mol/L}$), tranlylcypromine (a CYP2A6 inhibitor, 1 $\mu\text{mol/L}$), thiotepa (a CYP2B6 inhibitor, 20 $\mu\text{mol/L}$), trimethoprim (a CYP2C8 inhibitor, 100 $\mu\text{mol/L}$), sulfaphenazole (a CYP2C9 inhibitor, 50 $\mu\text{mol/L}$), (+)-*N*-3-benzyl nirvanol (a CYP2C19 inhibitor, 1 $\mu\text{mol/L}$), or quinidine (a CYP2D6 inhibitor, 5 $\mu\text{mol/L}$) did not inhibit the metabolism of istradefylline.

Microsomes expressing human CYP isoforms (CYP1A1, 1A2, 2A6, 2B6, 2C8, 2C9*1, 2C18, 2C19, 2D6*1, 2E1, 3A4, 3A5) were incubated with 1 $\mu\text{mol/L}$ of istradefylline at 37°C for 30 minutes. The following metabolite peaks were identified: M1 (4'-*O*-demethyl istradefylline) and eight other different metabolites detected from the microsomes expressing CYP1A1; M1, M2 (1- β -hydroxylated-4'-*O*-demethyl istradefylline), M8 (1- β -hydroxylated istradefylline), and five other different metabolites detected from the microsomes expressing CYP3A4; and M1, M8, and one other metabolite detected from the microsomes expressing CYP3A5. A weak peak for M1 was detected from the microsomes expressing CYP1A2, 2B6, 2C8, 2C18, and 2D6*1. The amounts of istradefylline that remained unchanged in the reaction mixture containing the microsomes expressing CYP1A1, 3A4, and 3A5 were 3.1%, 47.9%, and 65.0%, respectively, of those with control microsomes expressing no CYP.

Microsomes expressing human CYP1A1, 3A4, and 3A5 were added with 1 to 10,000 nmol/L of istradefylline and the enzyme kinetic parameters for the metabolism of istradefylline were determined. CYP1A1 displayed Michaelis-Menten kinetics with an intrinsic clearance of 42.6 $\mu\text{L/min/pmol}$ CYP1A1 and CYP3A4 and CYP3A5 showed autoactivation kinetics with maximum intrinsic clearance values of 1.17 $\mu\text{L/min/pmol}$ CYP3A4 and 0.572 $\mu\text{L/min/pmol}$ CYP3A5, respectively.

(b) Inhibition of CYP3A4/5 by istradefylline and M1 (Attached document 4.2.2.4-13, 4.2.2.4-15 to 4.2.2.4-16)

CYP3A4 inhibition was determined when istradefylline and the substrate were added simultaneously to human liver microsomes. Human liver microsomes were incubated with istradefylline or M1 (both 0.75-30 $\mu\text{mol/L}$) and testosterone (30 $\mu\text{mol/L}$) or nifedipine (10 $\mu\text{mol/L}$) in the presence of a reduced nicotinamide adenine dinucleotide phosphate (NADPH)-generating system at 37°C for 10 minutes (“coincubation”). Istradefylline and M1 inhibited CYP3A4/5-mediated testosterone 6- β -hydroxylation and nifedipine oxidation in a concentration-dependent manner and the IC_{50} values for the inhibition of testosterone 6- β -hydroxylation and nifedipine oxidation were ≥ 7.5 $\mu\text{mol/L}$ and 6.24 $\mu\text{mol/L}$, respectively, for istradefylline and 1.76 and 1.51 $\mu\text{mol/L}$, respectively, for M1.

CYP3A4 inhibition was determined when human liver microsomes were preincubated with istradefylline. Human liver microsomes were preincubated with istradefylline or M1 (both 0.75, 3, 7.5, and 30 $\mu\text{mol/L}$) in the presence of an NADPH-generating system at 37°C for 20 minutes and then testosterone (30 $\mu\text{mol/L}$) or nifedipine (10 $\mu\text{mol/L}$) was added and incubation was continued at 37°C for 10 minutes (“preincubation”). The IC_{50} values for the inhibition of testosterone 6- β -hydroxylation and nifedipine oxidation were < 0.75 $\mu\text{mol/L}$ and 0.871 $\mu\text{mol/L}$, respectively, for istradefylline and both < 0.75 $\mu\text{mol/L}$ for M1.

CYP3A4 inhibition was determined when microsomes expressing CYP3A4 were preincubated with istradefylline. Microsomes expressing CYP3A4 were preincubated with ^{14}C -istradefylline (5 $\mu\text{mol/L}$) at 37°C for 60 minutes in the absence or presence of an NADPH-generating system and then the preincubation mixture was added to a solution containing testosterone (250 $\mu\text{mol/L}$) and NADPH and incubated at 37°C for 10 minutes. Testosterone 6- β -hydroxylase activities were 56.7 and 3.57 nmol/min/nmol CYP3A4, respectively, showing that the activity was reduced in the presence of an NADPH-generating system and testosterone 6- β -hydroxylation in the microsomes expressing CYP3A4 was inhibited by preincubation with istradefylline in the presence of an NADPH-generating system.

CYP3A4 inhibition was determined when istradefylline and the substrate were added simultaneously to cryopreserved human hepatocytes or istradefylline was preincubated with cryopreserved human hepatocytes. When two lots of cryopreserved human hepatocytes were incubated with testosterone (250 $\mu\text{mol/L}$) and istradefylline (0.1-10 $\mu\text{mol/L}$) at 37°C for 20 minutes, testosterone 6- β -hydroxylase activities were 74.1% to 103% of those in the absence of istradefylline. In the presence of a CYP3A4 inhibitor, ketoconazole (2 $\mu\text{mol/L}$) as a positive control, testosterone 6- β -hydroxylation was inhibited by about 85% to 90%. Cryopreserved human hepatocytes were preincubated with 10 $\mu\text{mol/L}$ of istradefylline at 37°C for 60 minutes and then testosterone (250 $\mu\text{mol/L}$) was added and incubation was continued at 37°C for 20 minutes. As a result, testosterone 6- β -hydroxylase activities in the two lots of hepatocytes were reduced to 40.2% and 24.1%, respectively, of those in the absence of istradefylline and testosterone 6- β -hydroxylation was inhibited by preincubation with istradefylline.

(c) Inhibition of other CYP isoforms (Attached document 4.2.2.4-13 to 4.2.2.4-14)

The inhibitory effects of istradefylline and M1 on CYP isoform-specific reactions (ethoxyresorufin O-deethylation [CYP1A1/2], coumarin 7-hydroxylation [CYP2A6], bupropion hydroxylation [CYP2B6], tolbutamide methylhydroxylation [CYP2C8/9], *S*-mephenytoin 4'-hydroxylation [CYP2C19], bufuralol 1'-hydroxylation [CYP2D6], chlorzoxazone 6-hydroxylation [CYP2E1]) were determined in human liver microsomes under coincubation and preincubation conditions. Istradefylline or M1 had little effect on these reactions under either condition.

(d) CYP induction by istradefylline (Attached document 4.2.2.4-12)

HepaRG cells were exposed to istradefylline (0.1, 1, 10 µmol/L), omeprazole (100 µmol/L), or vehicle for 73 hours and then incubated with phenacetin for 1 hour. Compared with vehicle, omeprazole produced a 46.3-fold increase in CYP1A2 activity quantified by measuring the formation of acetaminophen while CYP1A2 activity in cells exposed to istradefylline were 0.927- to 1.22-fold that observed in the vehicle control.

HepaRG cells were exposed to istradefylline (0.1, 1, 10 µmol/L), rifampicin (10 µmol/L), or vehicle for 73 hours and then incubated with testosterone for 1 hour. Compared with vehicle, rifampicin produced a 18.4-fold increase in CYP3A4 activity, represented by testosterone 6-β-hydroxylation while CYP3A4 activity in cells exposed to istradefylline was 0.654- to 1.62-fold that observed in the vehicle control.

4.(ii).A.(2) Studies in healthy adult subjects

4.(ii).A.(2).1 Japanese single-dose study (Study 6002-9601, Attached document 5.3.3.1-1 to 5.3.3.1-2, 5.3.3.1-4)

Following administration of single oral doses of 10, 25, 50, 100, 150, and 200 mg of istradefylline in 36 Japanese healthy adult male subjects ($n = 6/\text{group}$) under fasted conditions, the C_{max} values of plasma istradefylline were 43.0 ± 17.7 , 113.1 ± 50.4 , 166.4 ± 53.0 , 247.4 ± 72.7 , 260.4 ± 62.9 , and 342.3 ± 78.2 ng/mL, respectively, the AUC_{0-t} values were 275.6 ± 212.0 , 2387.7 ± 1322.5 , 4539.8 ± 1623.9 , 7945.7 ± 2699.4 , 8479.2 ± 1352.5 , and $13,227.4 \pm 3653.1$ ng·h/mL, respectively, and the median t_{max} ranged from 2 to 4 hours across the dose groups. The $t_{1/2}$ values in the 25, 50, 100, and 150 mg groups were 34.4 ± 11.9 , 41.6 ± 28.9 , 44.3 ± 17.6 , and 39.3 ± 14.5 hours, respectively. Plasma concentrations of istradefylline during the elimination phase were less than the lower limit of quantitation in most subjects of the 10 mg group and the $t_{1/2}$ was calculable for 1 subject only and the $t_{1/2}$ in this subject was 31.3 hours. In the 200 mg group, the decline in istradefylline plasma concentration was triphasic and the $t_{1/2}$ was not calculable due to the lack of adequate elimination phase concentration-time data up to 72 hours post-dose. The plasma concentrations of M1 at and around the t_{max} of plasma istradefylline were about 1/30 of the plasma concentrations of istradefylline at all dose levels.

The 72-hour cumulative urinary excretion of istradefylline and M1 accounted for 4.60×10^{-3} to $10.4 \times 10^{-3}\%$ and 9.94×10^{-3} to $22.3 \times 10^{-3}\%$ of the administered dose, respectively.

4.(ii).A.(2).2) Japanese multiple-dose studies

(a) Study 6002-9703 (Attached document 5.3.3.1-5)

Istradefylline 20 mg was administered once daily after a meal for 14 days to 9 Japanese healthy adult male subjects. The median t_{\max} values of plasma istradefylline on Days 1 and 14 were both 2.00 hours, the C_{\max} values were 143.2 ± 18.5 and 230.9 ± 74.9 ng/mL, respectively, the areas under the plasma concentration-time curve from 0 to 24 hours (AUC_{0-24}) were 1209 ± 188 and 3249 ± 1516 ng·h/mL, respectively, and the $t_{1/2}$ of plasma istradefylline on Day 14 was 72.1 ± 47.4 hours. The plasma concentrations of M1 were less than the lower limit of quantitation at all timepoints in 6 subjects and quantifiable but ≤ 10 ng/mL at several timepoints in the remaining three subjects. The plasma levels of the *cis*-istradefylline were less than the lower limit of quantitation in all subjects.

The mean cumulative urinary excretion of istradefylline and M1 expressed as percent of total dose excreted during the period between the first dose and 96 hours after the last dose was $11.4 \times 10^{-3}\%$ and $4.4 \times 10^{-3}\%$, respectively, and the urine concentrations of *cis*-istradefylline were less than the lower limit of quantitation in all subjects.

(b) Study 6002-0104 (Attached document 5.3.3.1-8)

Pharmacokinetic parameters following once-daily administration of 20, 40, and 80 mg of istradefylline after a meal for 14 days to 27 Japanese healthy adult male subjects ($n = 9/\text{group}$) are shown in Table 3.

Table 3. Multiple-dose pharmacokinetic parameters of istradefylline

Dose	Day	t_{\max} (h)	C_{\max} (ng/mL)	C_{trough} (ng/mL)	AUC_{0-24} (ng·h/mL)	$t_{1/2}$ (h)
20 mg (n = 9)	1	2.00 [1.00-6.00]	149.2 ± 25.3	33.4 ± 11.5	1319 ± 335	—
	14	4.00 [2.00-4.00]	257.5 ± 88.0	154.6 ± 59.4	4406 ± 1598	75.0 ± 32.0
40 mg (n = 9)	1	2.00 [1.00-4.00]	257.3 ± 38.7	67.2 ± 20.3	2638 ± 616	—
	14	2.00 [0.50-4.00]	458.7 ± 117.4	284.7 ± 66.6	7925 ± 2047	59.1 ± 27.0
80 mg (n = 9)	1	2.00 [2.00-4.00]	391.2 ± 120.0	105.2 ± 38.0	3966 ± 1264	—
	14	2.00 [2.00-4.00]	857.3 ± 180.5	502.1 ± 136.2	$14,318 \pm 3023$	51.1 ± 25.0

Mean \pm SD, t_{\max} : Median [Min-Max]

C_{trough} : plasma trough concentration

M1 in plasma was quantifiable in 1 subject on Day 1 and 7 subjects on Day 14 in the 20 mg group and all subjects on both Days 1 and 14 in the 40 and 80 mg groups and the C_{\max} and AUC_{0-24} of M1 on Day 14 were about 1/35 to 1/40 and about 1/40 to 1/50 of those of istradefylline, respectively.

4.(ii).A.(2).3) Mass balance study (Study 6002-US-010, Attached document 5.3.3.1-11 [Reference data])

Following a single dose of 40 mg of ^{14}C -istradefylline administered as suspension in 6 foreign healthy adult

male subjects, $38.9 \pm 10.7\%$ and $48.0 \pm 13.4\%$ of the administered radioactivity were excreted in urine and feces, respectively, up to 18 days post-dose. The t_{\max} of plasma radioactivity was 1.50 ± 0.55 hours and about 80% and 60% of total radioactivity in plasma were present as istradefylline at 2 and 24 hours post-dose, respectively, and M5, M4, M8, and M1 were detected at 2 hours post-dose and M11 (a glucuronide conjugate of hydrogenated M1), M12 (a monoglucuronide conjugate of M10), M5, M4, and M8 were detected at 24 hours post-dose. In urine up to 168 hours post-dose, M11, M12, and M15 (a monosulfate conjugate of M14) combined accounted for 15.72% of the administered radioactivity, M16 (a sulfate conjugate of 1-deethyl-4'-*O*-demethyl istradefylline) accounted for 1.93% of the administered radioactivity, M5 and M19 (a monosulfate conjugate of M10) combined accounted for 4.67% of the administered radioactivity, and M4 accounted for 1.96% of the administered radioactivity, but istradefylline was not detected. In feces up to 168 hours post-dose, istradefylline was predominant, accounting for about 12% of the administered radioactivity, followed by M10 (3',4'-*O*-didemethyl-hydrogenated istradefylline) (5.77%), M14 (1- β -hydroxylated-3',4'-*O*-didemethyl-hydrogenated istradefylline) (3.94%), M13 (1-deethyl-3',4'-*O*-didemethyl-hydrogenated istradefylline) (3.08%), M17 (1-carboxymethyl istradefylline) (2.76%), and M18 (3',4'-*O*-didemethyl-1-carboxymethyl-hydrogenated istradefylline) (2.70%).

4.(ii).A.(3) Pharmacokinetics in patients with Parkinson's disease (Attached document 5.3.3.5-1)

Using plasma concentration data from Study 6002-0406, Study 6002-0608, Study 6002-009, and Study 6002-010 in which levodopa-treated Parkinson's disease patients with motor complications received 20 and 40 mg of istradefylline, a population pharmacokinetic (PPK) analysis was performed. Blood samples were collected at the end of Weeks 2, 4, and 12 (or at withdrawal) in Study 6002-0406, Study 6002-0608, and Study 6002-009 and at the end of Weeks 4 and 8 in Study 6002-010. The PPK analysis data set included 636 patients (277 male patients, 359 female patients) (2144 sampling points). A two-compartment model with first-order absorption was selected to describe the PPK of istradefylline and as the PPK parameters, apparent clearance (CL/F), apparent volume of distribution of central compartment (V_1/F), apparent intercompartmental clearance (Q/F), apparent volume of distribution of peripheral compartment (V_2/F), and absorption rate constant (k_a) were estimated. The distribution of the main background factors among the patients included in the PPK analysis was as follows: gender, 277 male patients and 359 female patients; age, 65.1 ± 8.4 years (mean \pm SD); body weight, 55.24 ± 11.62 kg; aspartate aminotransferase (AST), 22.8 ± 15.0 U/L; alanine aminotransferase (ALT), 16.3 ± 10.4 U/L; and creatinine clearance (C_{cr}), 79.95 ± 23.00 mL/min. The effects of these intrinsic factors as covariates were examined. The effects of the extrinsic factors of smoking status (38 smokers, 598 non-smokers) and food intake within 3 hours prior to taking the drug (yes, 578 patients; no, 58 patients) as covariates were examined.

The significant covariate was an effect of gender on CL/F and there was no significant covariate effect on V_1/F . The typical PPK parameters of istradefylline estimated from the final model were CL/F (L/h) = $4.98 \times 0.884^{\text{gender}}$ (gender, 0 for men; 1 for women), $V_1/F = 53.5 \pm 4.9$ L, Q/F = 24.9 L/h, $V_2/F = 494$ L, and $k_a = 0.254 \text{ h}^{-1}$. Interindividual variability was included in CL/F and V_1/F only and the coefficient of variation (CV) for CL/F and V_2/F were 33.9% and 98.7%, respectively.

4.(ii).A.(4) Intrinsic factor pharmacokinetic studies

4.(ii).A.(4).1) Single-dose study in elderly and non-elderly subjects (Study 6002-0205, Attached document 5.3.3.3-1)

Following administration of a single oral dose of 40 mg of istradefylline in 9 Japanese healthy non-elderly male subjects (20-33 years of age) and 9 Japanese healthy elderly male subjects (67-82 years of age) under fasted conditions, the median t_{\max} values of plasma istradefylline were 2.00 and 2.00 hours, respectively, the C_{\max} values were 172.6 ± 30.3 and 170.0 ± 47.7 ng/mL, respectively, and the AUC_{0-t} values were 4411 ± 2272 and 5469 ± 2991 ng·h/mL, respectively. The geometric mean ratios (elderly/non-elderly) of C_{\max} and AUC_{0-t} [95% CI] were 0.965 [0.759-1.225] and 1.232 [0.724-2.098], respectively.

4.(ii).A.(4).2) Pharmacokinetic study in subjects with renal impairment (Study 6002-US-015, Attached document 5.3.3.3-3)

Foreign healthy adult subjects, foreign subjects with renal impairment ($CL_{cr} < 30$ mL/min based on Cockcroft-Gault formula), and their age-matched foreign healthy adult subjects (6 subjects each) received a single oral dose of 40 mg of istradefylline under fasted conditions. The median t_{\max} values of plasma istradefylline were 2.50, 3.50, and 2.50 hours, respectively, the C_{\max} values were 215.9 ± 92.6 , 207.6 ± 83.1 , and 242.8 ± 43.8 ng/mL, respectively, the AUC_{0-t} values were $10,121 \pm 4211$, 9224 ± 4508 , and $10,834 \pm 3709$ ng·h/mL, respectively, and the $t_{1/2}$ values were 117.2 ± 70.2 , 116.9 ± 45.1 , and 94.8 ± 46.5 hours, respectively. The geometric mean ratios (subjects with renal impairment/their age-matched healthy adult subjects) of C_{\max} and AUC_t [90% CI] were 0.797 [0.508-1.250] and 0.821 [0.495-1.362], respectively. The plasma protein binding was $97.6 \pm 1.2\%$ in healthy adult subjects, $97.2 \pm 1.7\%$ in subjects with renal impairment, and $97.7 \pm 1.6\%$ in their age-matched healthy adult subjects.

4.(ii).A.(4).3) Pharmacokinetic study in subjects with hepatic impairment (Study 6002-US-016, Attached document 5.3.3.3-4)

Istradefylline 40 mg was orally administered once daily after a meal for 14 days in foreign subjects with hepatic impairment (moderate hepatic impairment based on Child-Pugh classification) and their age-matched foreign healthy adult subjects (7 subjects each). The median t_{\max} values of plasma istradefylline were 3.00 and 3.00 hours, respectively, the C_{\max} values were 506.1 ± 140.0 and 492.8 ± 125.0 ng/mL, respectively, the AUC_{0-24} values were 8431 ± 2538 and 9181 ± 2382 ng·h/mL, respectively, and the $t_{1/2}$ values were 287.6 ± 204.8 and 118.0 ± 17.5 hours, respectively. While a steady state was nearly reached after 14 days of dosing in healthy adult subjects, the $t_{1/2}$ was prolonged and a steady state was not reached after 14 days of dosing in subjects with hepatic impairment. The plasma protein binding was $97.3 \pm 2.11\%$ in subjects with hepatic impairment and $98.2 \pm 0.50\%$ in their age-matched healthy adult subjects.

4.(ii).A.(5) Extrinsic factor pharmacokinetic studies

4.(ii).A.(5).1) Effect of istradefylline on the pharmacokinetics of levodopa/carbidopa (Study 6002-US-

009, Attached document 5.3.3.4-1)

Twenty-four foreign healthy adult subjects received a single dose of 200/50 mg levodopa/carbidopa under fasted conditions (Day 1) followed by 80 mg istradefylline once daily after a meal for 13 days (Days 2-14) and then a single dose of 80 mg istradefylline in combination with 200/50 mg levodopa/carbidopa under fasted conditions on Day 15. The geometric mean ratios (levodopa+istradefylline/levodopa) of C_{\max} and the area under the plasma concentration-time curve from 0 to 12 hours (AUC_{0-12}) of levodopa [90% CI] were 1.12 [1.01-1.24] and 1.06 [1.01-1.10], respectively, and the geometric mean ratios of C_{\max} and AUC_{0-12} of carbidopa [90% CI] were 1.08 [0.99-1.18] and 1.03 [0.96-1.10], respectively.

4.(ii).A.(5).2) Effect of istradefylline on the pharmacokinetics of midazolam (Study 6002-US-008, Attached document 5.3.3.4-2)

Sixteen foreign healthy adult male subjects received a single oral dose of 10 mg midazolam after a meal (Day 1) followed 3 days later by istradefylline 80 mg orally once daily after a meal for 15 days (Days 4-18) with a single dose of 10 mg midazolam after a meal on Day 17. The geometric mean ratios (midazolam+istradefylline/midazolam) of C_{\max} and the area under the plasma concentration-time curve from zero to infinity ($AUC_{0-\infty}$) of midazolam [90% CI] were 1.61 [1.32-1.96] and 2.41 [2.13-2.73], respectively.

4.(ii).A.(5).3) Effect of istradefylline on the pharmacokinetics of atorvastatin (Study 6002-US-020, Attached document 5.3.3.4-3)

Sixteen foreign healthy adult male subjects received a single oral dose of 40 mg atorvastatin after a meal (Day 1) followed 4 days later by 40 mg istradefylline orally once daily after a meal for 17 days (Days 5-21) with a single dose of 40 mg atorvastatin after a meal on Day 18. The geometric mean ratios (atorvastatin+istradefylline/atorvastatin) of C_{\max} and $AUC_{0-\infty}$ of atorvastatin [90% CI] were 1.53 [1.34-1.75] and 1.54 [1.37-1.72], respectively, the geometric mean ratios of C_{\max} and $AUC_{0-\infty}$ of orthohydroxy atorvastatin [90% CI] were 0.95 [0.82-1.10] and 1.18 [1.07-1.30], respectively, and the geometric mean ratios of C_{\max} and $AUC_{0-\infty}$ of parahydroxy atorvastatin [90% CI] were 0.82 [0.64-1.06] and 1.06 [0.86-1.31], respectively.

4.(ii).A.(5).4) Effect of istradefylline on the pharmacokinetics of digoxin (Study 6002-US-026, Attached document 5.3.3.4-4)

A drug-drug interaction study was conducted in 24 foreign healthy adult male subjects. Each subject received a single oral dose of 0.4 mg digoxin under fasted conditions (Day 1) followed 14 days later by 40 mg istradefylline once daily under fasted conditions for 20 days (Days 15-34) and then a single dose of 40 mg istradefylline in combination with 0.4 mg digoxin under fasted conditions (Day 35). The geometric mean ratios (digoxin+istradefylline/digoxin) of C_{\max} , $AUC_{0-\infty}$, and renal clearance (CL_R) of digoxin [90% CI] were 1.33 [1.16-1.52], 1.21 [1.11-1.31], and 0.84 [0.76-0.92], respectively.

4.(ii).A.(5).5) Effect of ketoconazole on the pharmacokinetics of istradefylline (Study 6002-US-008,

Attached document 5.3.3.4-2)

Eighteen foreign healthy adult male subjects received a single oral dose of 40 mg istradefylline after a meal (Day 1) followed 14 days later by 200 mg ketoconazole orally twice daily for 4 days (Days 15-18). Subjects continued to receive 200 mg ketoconazole once daily for 7 days (Days 19-25) with a single dose of 40 mg istradefylline after a meal on Day 19. The geometric mean ratios (istradefylline+ketoconazole/istradefylline) of C_{\max} , $AUC_{0-\infty}$, and $t_{1/2}$ of istradefylline [90% CI] were 0.99 [0.86-1.14], 2.47 [1.92-3.18], and 1.87 [0.41-3.33], respectively.

4.(ii).A.(5).6 Effect of smoking on the pharmacokinetics of istradefylline (Study 6002-US-016, Attached document 5.3.3.3-4)

Istradefylline 40 mg was orally administered once daily after a meal for 14 days to foreign subjects with hepatic impairment who were smokers (moderate hepatic impairment based on Child-Pugh classification), their age-matched foreign healthy adult subjects who were smokers, foreign subjects with hepatic impairment who were non-smokers (moderate hepatic impairment based on Child-Pugh classification), and their age-matched foreign healthy adult subjects who were non-smokers (7 subjects each). The geometric mean ratios (smokers/non-smokers) of C_{\max} and AUC_{0-24} of istradefylline [90% CI] were 0.79 [0.54-1.17] and 0.58 [0.38-0.91], respectively, in healthy adult subjects, and 0.72 [0.49-1.06] and 0.64 [0.41-0.99], respectively, in subjects with hepatic impairment.

4.(ii).A.(6) QT/QTc study (Study 6002-US-024, Attached document 5.3.5.4-1)

A randomized, parallel-group study was conducted in 176 foreign healthy adult subjects (88 men and 88 women, 44 subjects each in the istradefylline 40 mg, istradefylline 160 mg, placebo, and moxifloxacin groups) to evaluate the effects of istradefylline on QTc interval. Istradefylline 40 mg, istradefylline 160 mg, placebo, or a positive control (moxifloxacin 400 mg on Days 1 and 14 and placebo on Days 2-13) was orally administered once daily for 14 days. Istradefylline and placebo were given in a double-blind fashion and moxifloxacin was given in a single-blind fashion.

On Day 14 after 14 days of administration of 40 mg and 160 mg of istradefylline, the median t_{\max} values were 3.00 and 4.00 hours, respectively, the C_{\max} values were 518.4 ± 171.2 and 1556.9 ± 516.8 ng/mL, respectively, and the AUC_{0-24} values were 9228 ± 3233 and $29,279 \pm 9263$ ng·h/mL, respectively.

The largest time-matched baseline-adjusted least-squares mean differences in QTcI between istradefylline 40 mg and placebo and between istradefylline 160 mg and placebo [90% CI] were 2.17 [-4.92 to 9.25] msec at 4 hours post-dose and 2.84 [-4.60 to 10.27] msec at 6 hours post-dose, respectively. The largest time-matched baseline-adjusted least-squares mean difference in QTcI between moxifloxacin and placebo [90% CI] was 11.50 [4.62-18.37] msec at 3 hours post-dose.

4.(ii).B Outline of the review by PMDA

4.(ii).B.(1) Pharmacokinetics in healthy adult subjects

At higher doses in a single-dose study in healthy adult subjects, the decline in istradefylline plasma concentration was multiphasic and the C_{\max} tended to level off. PMDA asked the applicant to explain their causes.

The applicant responded as follows:

In a single-dose study (Study 6002-9601), the decline in istradefylline plasma concentration was biphasic or triphasic at ≥ 12 hours post-dose at doses ≥ 50 mg and especially at the highest dose of 200 mg, it was multiphasic up to 48 hours post-dose. Such a multiphasic elimination pattern was also observed 24 to 32 hours following single oral administration of high-dose istradefylline in dogs under fasted conditions. The multiphasic elimination pattern in dogs is considered due to the dissolution and absorption of istradefylline remaining in the gastrointestinal tract over time and is not presumed to be caused by enterohepatic circulation because the unchanged drug was not detected in the bile in a rat study. In Study 6002-9601, subjects were fasted from 12 hours before dosing and the multiphasic decline in plasma concentrations observed at doses ≥ 50 mg are considered due to absorption of istradefylline remaining in the gastrointestinal tract, as in the case of dogs. Since the multiphasic decline was less apparent at 50 mg compared with higher doses in Study 6002-9601 and the absorption rate of istradefylline after oral administration of 40 mg under fasted conditions was high at about 90% in a mass balance study (Study 6002-US-010), it is inferred that a multiphasic decline in plasma concentrations of istradefylline does not occur over the dose range of 20 to 40 mg.

In Study 6002-9601, following single doses of 10, 25, 50, 100, 150, and 200 mg of istradefylline in healthy adult male subjects, the C_{\max} increased dose-proportionally over the dose range of 10 to 50 mg, but leveled off, showing nonlinearity at doses higher than 50 mg. It seemed that no reliable AUC_{0-t} data at 10 mg were available because plasma concentrations were quantifiable only at a few timepoints in most subjects. However, as the AUC_{0-t} was linear over the dose range of 25 to 200 mg, it is considered that the C_{\max} leveled off because dissolution was a rate-limiting step for the absorption of istradefylline.

PMDA considers as follows:

Since non-clinical data indicate that the enterohepatic circulation of istradefylline is unlikely, as the applicant explained, the cause for the multiphasic elimination profile or non-linear C_{\max} at higher doses may be the low solubility of istradefylline. In a mass balance study in which istradefylline was administered at the maximum recommended clinical dose of 40 mg, most of the administered radioactivity was excreted as metabolites and a small percentage of the administered radioactivity was excreted as istradefylline, indicating that istradefylline is well-absorbed. In single-dose and multiple-dose studies, the C_{\max} and AUC increased almost dose-proportionally over the dose range up to 40 mg. Therefore, the low solubility of istradefylline should not affect the exposure of istradefylline within the clinical dose range. Administration of 20 mg of istradefylline to humans under fed conditions resulted in a 10% to 20% increase in exposure as compared with under fasted conditions and it has been demonstrated that there are no major differences in food effect between 20 mg and 50 mg. Thus, patients treated with clinical doses of istradefylline are unlikely to

experience extreme increases in plasma concentrations, irrespective of the dosing conditions that would affect solubility, e.g. the amount and content of food, and the multiphasic decline in plasma concentrations of istradefylline or non-linear pharmacokinetics observed at doses higher than the recommended clinical doses is unlikely to be clinically relevant.

4.(ii).B.(2) Pharmacokinetics in subjects with hepatic impairment

The applicant explained about a pharmacokinetic study in subjects with hepatic impairment (Study 6002-US-016) as follows:

After 14-day administration of istradefylline, the $t_{1/2}$ was approximately 288 hours and the fraction of steady-state (f_{ss}) calculated from the $t_{1/2}$ value was 0.62 in subjects with hepatic impairment. On the other hand, the $t_{1/2}$ was 118 hours and the f_{ss} was 0.86 in healthy adult subjects. Since the f_{ss} was different between subjects with hepatic impairment and healthy adult subjects, it was considered inappropriate to compare the pharmacokinetics of istradefylline based on the C_{max} and AUC_{0-24} values on Day 14. Based on the accumulation index at steady state estimated from the $t_{1/2}$ value after 14-day administration in subjects with hepatic impairment and the ratio of the C_{max} on Day 14 to the C_{trough} during multiple-dose administration in subjects with hepatic impairment, the steady-state C_{max} and C_{trough} in subjects with hepatic impairment were estimated to be 1548 and 968 ng/mL, respectively. In the case of compounds with a long $t_{1/2}$ like istradefylline, the mean blood concentration at steady state was considered to approximate the mean of the C_{max} and C_{trough} values and the steady-state AUC_{0-24} was estimated to be 30,192 ng·h/mL. Based on the above, the steady-state C_{max} and AUC_{0-24} values in subjects with hepatic impairment are estimated to be roughly 3-fold those in healthy adult subjects.

PMDA considers as follows:

Since *in vitro* studies showed that istradefylline is metabolized by the liver and a mass balance study indicated that istradefylline is primarily eliminated by liver metabolism, it is necessary to be careful about the effect of hepatic impairment on the pharmacokinetics of istradefylline. As the applicant explained that a pharmacokinetic study in subjects with hepatic impairment failed to evaluate the steady-state pharmacokinetics of istradefylline, whether istradefylline should be used in patients with hepatic impairment and whether dose adjustment is required will be determined, taking also account of safety data [see “4.(iii) Summary of clinical efficacy and safety”].

4.(ii).B.(3) Effects of extrinsic factors on pharmacokinetics

1) Interactions with levodopa/carbidopa

Istradefylline is always used in combination with levodopa preparations. Istradefylline has been shown to have a small effect on the pharmacokinetics of levodopa/carbidopa in an istradefylline drug interaction study with levodopa/carbidopa, while the effect of levodopa/carbidopa on the pharmacokinetics of istradefylline has not been evaluated in this study. PMDA asked the applicant to explain drug-drug interactions when istradefylline and levodopa/carbidopa are administered in combination.

The applicant responded as follows:

There were no major differences between the plasma concentration-time profile after multiple-dose administration of istradefylline in healthy adult subjects and the plasma concentration-time profile in patients with Parkinson's disease on levodopa/carbidopa therapy estimated from the PPK model. It is considered that levodopa/carbidopa increased the systemic exposure of istradefylline in rats because of levodopa-induced inhibition of gastric emptying, resulting in increased absorption. Meanwhile, about 90% of the administered dose was absorbed in a mass balance study with 40 mg istradefylline, indicating that istradefylline is well-absorbed in the clinical dose range. Therefore, levodopa/carbidopa should have no effect on the systemic exposure of istradefylline in a clinical setting.

PMDA accepted the applicant's response and concluded that the effect of levodopa/carbidopa on the pharmacokinetics of istradefylline is unlikely to be clinically relevant.

2) Interactions with drugs that are CYP3A4 or P-glycoprotein substrates

As istradefylline increases the blood concentrations of drugs that are CYP3A4 or P-glycoprotein substrates, PMDA asked the applicant to consider whether any of the drugs that are CYP3A4 or P-glycoprotein substrates should be contraindicated for concomitant use with istradefylline.

The applicant responded as follows:

In drug-drug interaction studies, 80 mg of istradefylline increased the $AUC_{0-\infty}$ of midazolam (a substrate for CYP3A4) about 2.4-fold and 40 mg of istradefylline increased the $AUC_{0-\infty}$ of atorvastatin (a substrate for CYP3A4 and P-glycoprotein) about 1.5-fold and the $AUC_{0-\infty}$ of digoxin (a substrate for P-glycoprotein) about 1.2-fold. Drugs contraindicated for concomitant use with CYP3A4 substrate drugs include potent CYP3A4 inhibitors, i.e. HIV protease inhibitors, macrolide antibiotics, and azole antifungal drugs and telaprevir, efavirenz, delavirdine, cyclosporine, and tacrolimus and istradefylline is unlikely to increase the exposure of CYP3A4 substrate drugs to an extent similar to the effects of these CYP3A4 inhibitors. Drugs contraindicated for concomitant use with P-glycoprotein substrate drugs include itraconazole and cyclosporine and istradefylline is unlikely to increase the exposure of P-glycoprotein substrate drugs to an extent similar to the effect of itraconazole or cyclosporine. Therefore, there is no need to contraindicate concomitant use of istradefylline with drugs that are CYP3A4 or P-glycoprotein substrates. The results of istradefylline interaction studies will be presented in the package insert.

PMDA considers as follows:

Since istradefylline is not a potent CYP3A4 or P-glycoprotein inhibitor compared with drugs contraindicated for concomitant use with CYP3A4 or P-glycoprotein substrate drugs that have been approved in Japan, there is no need to contraindicate concomitant use of istradefylline with CYP3A4 or P-glycoprotein substrate drugs at present. Therefore, the applicant's responses (drugs that are CYP3A4 or P-glycoprotein substrates will be listed in the precautions for coadministration section of the package insert and the extent of increase in the plasma concentration of each drug observed in istradefylline drug interaction studies will be presented in the package insert) are appropriate. However, as the safety information on istradefylline when administered with CYP3A4 or P-glycoprotein substrate drugs in clinical studies is limited, it is necessary to collect safety

information on the concomitant use of istradefylline with CYP3A4 or P-glycoprotein substrate drugs via post-marketing surveillance etc. The precaution statement regarding the concomitant use of istradefylline with CYP3A4 or P-glycoprotein substrate drugs will be finalized, taking account of comments from the Expert Discussion.

3) Concomitant use with CYP3A4 inhibitors

Ketoconazole (a CYP3A4 inhibitor) increased the $AUC_{0-\infty}$ of istradefylline about 2.5-fold (Study 6002-US-008). PMDA asked the applicant to explain whether istradefylline can be coadministered with CYP3A4 inhibitors.

The applicant explained as follows:

In a long-term treatment study in patients with Parkinson's disease (Study 6002-010), 27 of 308 patients used concomitant CYP3A4 inhibitors and plasma concentrations after coadministration of istradefylline with CYP3A4 inhibitors were obtained from 3 patients treated with 20 mg and 3 patients treated with 40 mg among the 27 patients and the CYP3A4 inhibitors used included clarithromycin, glibenclamide, and fosfluconazole. The exposure of istradefylline (AUC_{0-24}) in patients on concomitant CYP3A4 inhibitors was estimated using the population parameters obtained from the PPK analysis. As a result, there were no major differences in the exposure between patients receiving istradefylline with and without CYP3A4 inhibitors. Adverse events occurring after coadministration of istradefylline with CYP3A4 inhibitors were analyzed. As a result, adverse events reported by at least 3 patients were nasopharyngitis (10 patients) and contusion (5 patients) and all of these events were non-serious and mild in severity and assessed as unrelated to istradefylline. Thus, there should be no safety concerns about concomitant use with CYP3A4 inhibitors.

PMDA considers as follows:

As the concomitant use of CYP3A4 inhibitors was prohibited in Japanese placebo-controlled studies (Study 6002-0406, Study 6002-0608, Study 6002-009), no istradefylline blood concentrations or safety data when istradefylline was administered with CYP3A4 inhibitors were obtained from these studies. Long-term treatment data raised no specific safety concerns for patients receiving istradefylline with CYP3A4 inhibitors compared with those without CYP3A4 inhibitors, but the data on concomitant use of istradefylline with CYP3A4 inhibitors are very limited. Ketoconazole increased the $AUC_{0-\infty}$ of istradefylline about 2.5-fold in a drug-drug interaction study and if istradefylline 40 mg is coadministered with CYP3A4 inhibitors, the exposure may exceed that at 80 mg, which is the maximum dose of istradefylline administered as multiple doses in Japanese subjects. As a supraclinical dose of istradefylline leads to an increased incidence of adverse events such as psychiatric disorders and nausea, it is necessary to list CYP3A4 inhibitors in the precautions for coadministration section of the package insert and the dose of istradefylline should not exceed 20 mg when istradefylline is used concomitantly with potent CYP3A4 inhibitors like ketoconazole. Although the applicant explained that based on the analysis using the plasma concentration data from the long-term treatment study, the plasma exposure of istradefylline in patients receiving istradefylline with CYP3A4 inhibitors is not different from that in those without CYP3A4 inhibitors, as plasma concentrations after coadministration of istradefylline with CYP3A4 inhibitors were obtained from the limited number of patients,

it is difficult to explain the effects of concomitant CYP3A4 inhibitors on the pharmacokinetics of istradefylline based on the long-term treatment data. The precaution statement regarding the concomitant use of istradefylline with CYP3A4 inhibitors will be finalized, taking account of comments from the Expert Discussion.

4) Effect of smoking

The applicant explained the effect of smoking on the pharmacokinetics of istradefylline as follows:

The plasma exposure of istradefylline was lower in smokers than in non-smokers (C_{\max} ratio, 79.3%; AUC_{0-24} ratio, 58.4%) and the $t_{1/2}$ tended to be shorter in smokers than in non-smokers. One of the primary metabolizing enzymes for istradefylline is CYP1A1 and CYP1A2 is also involved in the metabolism of istradefylline. It is considered that CYP1A1 and CYP1A2 induction caused by smoking increased istradefylline clearance and decreased the plasma exposure of istradefylline.

PMDA considers as follows:

There is inter- and intra-individual variability in the number of cigarettes smoked and it is difficult to recommend uniform istradefylline dose adjustment for smokers. As the plasma exposure of istradefylline is reduced in smokers, a safety issue is unlikely to arise. Since the relationship between plasma exposure and efficacy (a reduction in hours of OFF time) is undefined, no dose adjustment is required for smokers at present. However, as a study demonstrated a reduction in plasma exposure of istradefylline in smokers compared with non-smokers, the package insert should include a statement cautioning about the effect of smoking on the pharmacokinetics of istradefylline and then patients should also be advised about the effect of smoking. It is also necessary to collect information on the effect of smoking on the safety and efficacy of istradefylline via post-marketing surveillance etc.

4.(iii) Summary of clinical efficacy and safety

4.(iii).A Summary of the submitted data

As the evaluation data, the results from a total of 16 studies (5 Japanese clinical pharmacology studies, 2 Japanese phase II studies, 1 Japanese phase III study, 1 Japanese long-term treatment study, 7 foreign clinical pharmacology studies) were submitted. Additional data generated during the assessment period of the registration application (a BE study for different dosage form strengths) were submitted [see “4.(i) Summary of biopharmaceutic studies and associated analytical methods” and “4.(ii) Summary of clinical pharmacology studies” for BE and pharmacokinetic data].

4.(iii).A.(1) Japanese clinical pharmacology studies

4.(iii).A.(1).1 Food effect study with istradefylline in the final to-be-marketed dosage form (Study 6002-011, Attached document 5.3.1.1-1; Studied period, June to July 2011)

An open-label, two-way crossover study was conducted in 20 healthy adult male subjects at a single center in Japan to evaluate the food effect on the pharmacokinetics of istradefylline and the safety of istradefylline. Each subject received a single oral dose of 20 mg of istradefylline in the final to-be-marketed dosage form (the 20-mg tablet) under fasted and fed conditions (a ≥ 21 -day washout period). All of 20 subjects treated

with study drug completed the study.

An adverse event of creatine phosphokinase (CK) increased occurred in 1 subject, no deaths or serious adverse events were reported, and there were no noteworthy laboratory or vital sign changes.

4.(iii).A.(1).2) Single-dose study (Study 6002-9601, Attached document 5.3.3.1-1; Studied period, June 1996 to March 1997)

A single-blind study was conducted in 60 healthy adult male subjects (Steps 1-6, 8 subjects each [6 subjects receiving istradefylline, 2 subjects receiving placebo]; Step 7, 12 subjects) at a single center in Japan to evaluate the safety and pharmacokinetics of a single dose of istradefylline and the food effect on the safety and pharmacokinetics of istradefylline. A single oral dose of 10, 25, 50, 100, 150, or 200 mg of istradefylline or placebo was administered under fasted conditions (Steps 1-6) or a single oral dose of 50 mg of istradefylline was administered under fasted and fed conditions in a crossover fashion (a 2-week washout period) (Step 7). All of 60 subjects treated with study drug completed the study.

Adverse events based on subjective symptoms were reported by 5 subjects in the placebo group (headache in 2 subjects, somnolence in 2 subjects, vertigo in 1 subject, dizziness on standing up in 1 subject, generalised hot flushes in 1 subject), 1 subject in the 10 mg group (itchy sensation of forearms and back), 2 subjects in the 25 mg group (fuzzy head in 2 subjects), 3 subjects in the 50 mg group (poor sleep, floating feeling, hypoaesthesia of legs, nasal discharge, and sensation of closed nose, one subject each), 5 subjects in the 100 mg group (floating feeling in 2 subjects, generalised hot flushes in 1 subject, palpitations in 1 subject, strange sensation of the pharynx in 1 subject, strange sensation of the posterior neck in 1 subject, difficulty falling asleep in 1 subject), 5 subjects in the 150 mg group (facial hot flushes in 2 subjects, difficulty falling asleep in 2 subjects, generalised hot flushes in 1 subject, dizziness on standing up in 1 subject, hot feeling generalized in 1 subject, floating feeling in 1 subject), and 3 subjects in the 200 mg group (poor sleep, epigastric heaviness, pain pharynx, and increased salivation, one subject each) in Steps 1 to 6 and 4 subjects after fasted administration (difficulty falling asleep in 2 subjects, floating feeling in 1 subject, mental concentration decreased in 1 subject, cough in 1 subject) in Step 7. No deaths or serious adverse events were reported and there were no clinically relevant laboratory or vital sign abnormalities.

4.(iii).A.(1).3) Multiple-dose study (a) (Study 6002-9703, Attached document 5.3.3.1-5; Studied period, July to September 1998)

A single-blind study was conducted in 12 healthy adult male subjects (9 subjects in the istradefylline group, 3 subjects in the placebo group) at a single center in Japan to evaluate the safety and pharmacokinetics of multiple doses of istradefylline. Istradefylline 20 mg or placebo was orally administered once daily after a meal for 14 days. All of 12 subjects treated with study drug completed the study.

Adverse events reported include aldolase increased [100% (9 of 9 subjects) in the istradefylline group, 66.7% (2 of 3 subjects) in the placebo group], acid phosphatase increased [66.7% (6 of 9 subjects) in the istradefylline group, 66.7% (2 of 3 subjects) in the placebo group], a general sense of openness [22.2% (2 of

9 subjects) in the istradefylline group, 0% (0 of 3 subjects) in the placebo group], and talkativeness [11.1% (1 of 9 subjects) in the istradefylline group, 0% (0 of 3 subjects) in the placebo group]. No deaths or serious adverse events were reported. Other than the abnormal laboratory changes reported as adverse events, there were no clinically relevant laboratory or vital sign abnormalities.

4.(iii).A.(1).4 Multiple-dose study (b) (Study 6002-0104, Attached document 5.3.3.1-8; Studied period, April to August 2002)

A single-blind study was conducted in 36 healthy adult male subjects (12 subjects in each step [9 subjects receiving istradefylline, 3 subjects receiving placebo]) at a single center in Japan to evaluate the safety and pharmacokinetics of multiple doses of istradefylline. Istradefylline 20, 40, or 80 mg or placebo was orally administered once daily after a meal for 14 days. One of 36 subjects treated with study drug was withdrawn from the study for a personal reason.

Adverse events were reported by 1 subject in the 20 mg group (ALT increased, AST increased, γ -glutamyltransferase increased) and 2 subjects in the 80 mg group (blood corticotrophin increased in 2 subjects). No deaths or serious adverse events were reported. Other than the abnormal laboratory changes reported as adverse events, there were no clinically relevant laboratory or vital sign abnormalities.

4.(iii).A.(1).5 Pharmacokinetic study in elderly subjects (Study 6002-0205, Attached document 5.3.3.3-1; Studied period, March to April 2003)

An open-label study was conducted in 18 healthy adult male subjects (9 elderly subjects, 9 non-elderly subjects) at a single center in Japan to evaluate the safety and pharmacokinetics of istradefylline in healthy elderly male subjects. A single oral dose of 40 mg of istradefylline was administered under fasted conditions. All of 18 subjects treated with study drug completed the study.

Adverse events were reported by 1 elderly subject (acute tonsillitis NOS) and 2 non-elderly subjects (activated partial thromboplastin time prolonged in 1 subject, pharyngolaryngeal pain in 1 subject). No deaths or serious adverse events were reported. Other than the abnormal laboratory change reported as an adverse event, there were no clinically relevant laboratory or vital sign abnormalities.

4.(iii).A.(1).6 BE study for different dosage form strengths (Study 6002-012, Attached document 5.3.1.2-1; Studied period, August to September 2012)

An open-label, two-way crossover study was conducted in 30 healthy adult male subjects at a single center in Japan to determine the BE between the 10-mg tablet and the 20-mg tablet. A single oral dose of two 10-mg tablets of istradefylline or one 20-mg tablet of istradefylline was administered under fasted conditions in each period (a ≥ 12 -day washout period). All of 30 subjects treated with study drug completed the study.

Adverse events were reported by 2 subjects after administration of the 20-mg tablet (pharyngitis, gastroenteritis). No deaths or serious adverse events were reported.

There were no clinically relevant laboratory or vital sign abnormalities.

4.(iii).A.(2) Foreign clinical pharmacology studies

4.(iii).A.(2).1 Pharmacokinetic study in subjects with severe renal impairment (Study 6002-US-015, Attached document 5.3.3.3-3; Studied period, March to October 2005)

An open-label study was conducted in subjects with severe renal impairment ($\text{Ccr}^* < 30 \text{ mL/min}$), their age-, BMI-, and sex-matched healthy adult subjects (age, ± 10 years; BMI, $\pm 25\%$; sex, a 100% match; $\text{Ccr} > 80 \text{ mL/min}$), and healthy young adult subjects (Ccr , age-matched normal values) (6 subjects each) at 3 centers overseas to evaluate the effect of severe renal impairment on the pharmacokinetics and safety of istradefylline. A single oral dose of 40 mg of istradefylline was administered under fasted conditions. All of 18 subjects treated with study drug completed the study.

The incidence of adverse events was 50.0% (3 of 6 subjects) in subjects with severe renal impairment, 16.7% (1 of 6 subjects) in healthy adult subjects, and 50.0% (3 of 6 subjects) in healthy young adult subjects and adverse events reported by at least 2 subjects in any group were constipation [33.3% (2 of 6 subjects) in subjects with severe renal impairment, 0% (0 of 6 subjects) in healthy adult subjects, 0% (0 of 6 subjects) in healthy young adult subjects]. No deaths or serious adverse events were reported and there were no clinically meaningful laboratory or vital sign changes.

4.(iii).A.(2).2 Pharmacokinetic study in smokers and subjects with moderate hepatic impairment (Study 6002-US-016, Attached document 5.3.3.3-4; Studied period, May to December 2005)

An open-label study was conducted in smoking (at least 20 cigarettes per day on average) and non-smoking subjects with hepatic impairment (Child-Pugh B) and their age-, BMI-, and sex-matched smoking and non-smoking healthy adult subjects (age, ± 10 years; BMI $\pm 25\%$; sex, a 100% match) (7 subjects each) at 2 centers overseas to evaluate the effect of moderate hepatic impairment on the pharmacokinetics and safety of multiple doses of istradefylline in smokers and non-smokers. Istradefylline 40 mg was orally administered once daily after a meal for 14 days (under fasted condition on Day 14 only). All of 28 subjects treated with study drug completed the study.

The incidence of adverse events was 14.3% (1 of 7 subjects) in smoking subjects with hepatic impairment, 28.6% (2 of 7 subjects) in smoking healthy adult subjects, 85.7% (6 of 7 subjects) in non-smoking subjects with hepatic impairment, and 42.9% (3 of 7 subjects) in non-smoking healthy adult subjects and adverse events reported by at least 2 subjects in any group were constipation [0% (0 of 7 subjects), 28.6% (2 of 7 subjects), 14.3% (1 of 7 subjects), and 0% (0 of 7 subjects), respectively], diarrhoea [0% (0 of 7 subjects), 0% (0 of 7 subjects), 28.6% (2 of 7 subjects), and 14.3% (1 of 7 subjects), respectively], and headache [14.3% (1 of 7 subjects), 0% (0 of 7 subjects), 28.6% (2 of 7 subjects), and 0% (0 of 7 subjects), respectively]. No deaths or serious adverse events were reported and there were no clinically meaningful laboratory or vital sign changes.

* Calculated using the Cockcroft-Gault formula.

4.(iii).A.(2).3) Effect of istradefylline on the pharmacokinetics of levodopa/carbidopa (Study 6002-US-009, Attached document 5.3.3.4-1; Studied period, October to November 2002)

An open-label study was conducted in 24 healthy adult subjects at a single center overseas to evaluate the effect of istradefylline on the pharmacokinetics of levodopa/carbidopa. A single oral dose of 200/50 mg levodopa/carbidopa was administered under fasted conditions on Days 1 and 15 and 80 mg istradefylline was orally administered once daily after a meal on Days 2 to 15 (under fasted condition on Day 15 only). All of 24 subjects treated with study drug completed the study.

The incidence of adverse events was 50.0% (12 of 24 subjects). Adverse events with an incidence of $\geq 10\%$ were queasy (25.0% [6 of 24 subjects]), constipation (16.7% [4 of 24 subjects]), dizziness (16.7% [4 of 24 subjects]), vomiting NOS (12.5% [3 of 24 subjects]), and depressed mood (12.5% [3 of 24 subjects]). No deaths or serious adverse events were reported and there were no clinically meaningful laboratory or vital sign changes.

4.(iii).A.(2).4) Istradefylline drug interaction study with a CYP3A4 substrate or inhibitor (Study 6002-US-008, Attached document 5.3.3.4-2; Studied period, September to November 2002)

An open-label study was conducted in 35 healthy adult subjects (Cohort 1, 17 subjects; Cohort 2, 18 subjects) at a single center overseas to evaluate the potential for drug-drug interactions between istradefylline and a CYP3A4 substrate (midazolam) or a CYP3A4 inhibitor (ketoconazole). In Cohort 1, a single oral dose of 10 mg midazolam was administered after a meal on Days 1 and 17 and 80 mg istradefylline was orally administered once daily after a meal on Days 4 to 18. In Cohort 2, a single oral dose of 40 mg istradefylline was administered after a meal on Days 1 and 19 and 200 mg ketoconazole was orally administered twice daily after a meal on Days 15 to 18 and once daily after a meal on Days 19 to 25. One subject in Cohort 1 (the occurrence of adverse events) and 2 subjects in Cohort 2 (the occurrence of adverse events, subject's request) were withdrawn from the study.

The incidence of adverse events was 100% (17 of 17 subjects) in Cohort 1 and 66.7% (12 of 18 subjects) in Cohort 2. Adverse events reported by at least 10% of subjects in Cohort 1 were somnolence (100% [17 of 17 subjects]), insomnia (35.3% [6 of 17 subjects]), headache NOS (23.5% [4 of 17 subjects]), palpitations (17.6% [3 of 17 subjects]), back pain (17.6% [3 of 17 subjects]), dizziness postural (17.6% [3 of 17 subjects]), logorrhoea (17.6% [3 of 17 subjects]), dry eye NOS (11.8% [2 of 17 subjects]), vision blurred (11.8% [2 of 17 subjects]), queasy (11.8% [2 of 17 subjects]), fatigue (11.8% [2 of 17 subjects]), feeling of relaxation (11.8% [2 of 17 subjects]), dizziness (11.8% [2 of 17 subjects]), restlessness (11.8% [2 of 17 subjects]), sleep disorder NOS (11.8% [2 of 17 subjects]), vaginal discharge (11.8% [2 of 17 subjects]), pharyngolaryngeal pain (11.8% [2 of 17 subjects]), and flushing (11.8% [2 of 17 subjects]). Adverse events reported by at least 10% of subjects in Cohort 2 were headache NOS (38.9% [7 of 18 subjects]), queasy (33.3% [6 of 18

subjects]), abdominal pain NOS (16.7% [3 of 18 subjects]), sleep disorder NOS (16.7% [3 of 18 subjects]), ear pain (11.1% [2 of 18 subjects]), dyspepsia (11.1% [2 of 18 subjects]), fatigue (11.1% [2 of 18 subjects]), dizziness (11.1% [2 of 18 subjects]), dysmenorrhoea (11.1% [2 of 18 subjects]), and postnasal drip (11.1% [2 of 18 subjects]).

In either cohort, no deaths or serious adverse events were reported and there were no clinically meaningful laboratory or vital sign changes.

4.(iii).A.(2).5 Istradefylline drug interaction study with atorvastatin (Study 6002-US-020, Attached document 5.3.3.4-3; Studied period, November to December 2004)

A double-blind study was conducted in 20 healthy adult male subjects (16 subjects in the istradefylline group, 4 subjects in the placebo group) at a single center overseas to evaluate the potential for drug-drug interactions between istradefylline and atorvastatin. A single oral dose of 40 mg atorvastatin was administered after a meal on Days 1 and 18 and 40 mg istradefylline or placebo was orally administered once daily after a meal on Days 5 to 21. All 20 subjects completed the study.

The incidence of adverse events was 31.3% (5 of 16 subjects) in the istradefylline group and 50.0% (2 of 4 subjects) in the placebo group and adverse events reported by at least 2 subjects in either group were insomnia (3 subjects in the istradefylline group, 0 subjects in the placebo group) and headache (2 subjects in the istradefylline group, 2 subjects in the placebo group). No deaths or serious adverse events were reported and there were no clinically meaningful laboratory or vital sign changes.

4.(iii).A.(2).6 Istradefylline drug interaction study with digoxin (Study 6002-US-026, Attached document 5.3.3.4-4; Studied period, August to September 2007)

An open-label study was conducted in 24 healthy adult male subjects at a single center overseas to evaluate the potential for drug-drug interactions between istradefylline and digoxin. A single oral dose of 0.4 mg digoxin was administered under fasted conditions on Days 1 and 35 and 40 mg istradefylline was orally administered once daily under fasted conditions on Days 15 to 35. Three subjects (protocol deviations [2 subjects], consent withdrawal [1 subject]) were withdrawn from the study.

The incidence of adverse events was 62.5% (15 of 24 subjects) and adverse events with an incidence of $\geq 10\%$ were blood CK increased (29.2% [7 of 24 subjects]) and headache (12.5% [3 of 24 subjects]). No deaths or serious adverse events were reported. Other than blood bilirubin increased (2 subjects), lipase increased (1 subject), and transaminases increased (1 subject) reported as adverse events, there were no clinically meaningful laboratory or vital sign changes.

4.(iii).A.(2).7 Thorough QTc study (Study 6002-US-024, Attached document 5.3.5.4-1; Studied period, August to October 2005)

A double-blind study was conducted in 176 healthy adult subjects (44 subjects per group) at a single center overseas to evaluate the effects of istradefylline on the QT (QTc) interval on ECG. Istradefylline 40 mg,

istradefylline 160 mg, or placebo was orally administered once daily after a meal for 14 days or 400 mg moxifloxacin was orally administered after a meal on Days 1 and 14.

All of 176 subjects were treated with study drug and included in the safety analysis set. Four subjects (1 subject in the istradefylline 40 mg group, 1 subject in the istradefylline 160 mg group, 2 subjects in the placebo group) were withdrawn from the study due to consent withdrawal.

The incidence of adverse events was 13.6% (6 of 44 subjects) in the istradefylline 40 mg group, 0% (0 of 44 subjects) in the istradefylline 160 mg group, 9.1% (4 of 44 subjects) in the moxifloxacin group, and 6.8% (3 of 44 subjects) in the placebo group. Adverse events reported by at least 2 subjects in any group were headache [6.8% (3 of 44 subjects), 0% (0 of 44 subjects), 4.5% (2 of 44 subjects), and 0% (0 of 44 subjects), respectively], back pain [2.3% (1 of 44 subjects), 0% (0 of 44 subjects), 0% (0 of 44 subjects), and 4.5% (2 of 44 subjects), respectively], and anxiety [4.5% (2 of 44 subjects), 0% (0 of 44 subjects), 0% (0 of 44 subjects), and 0% (0 of 44 subjects), respectively]. No deaths or serious adverse events were reported [see “4.(ii) Summary of clinical pharmacology studies” for QT interval data].

4.(iii).A.(3) Japanese clinical studies in patients with Parkinson’s disease

4.(iii).A.(3).1 Japanese early phase II study (Study 6002-0406, Attached document 5.3.5.1-1; Studied period, April 2005 to March 2006)

A randomized, double-blind, parallel-group, comparative study was conducted at 13 centers in Japan to evaluate the efficacy and safety of istradefylline in levodopa-treated Parkinson’s disease patients with motor complications (Target number of subjects, 25 subjects per group, a total of 75 subjects).

A 2-week screening period was followed by a 12-week double-blind treatment period in which 20 or 40 mg of istradefylline or placebo was orally administered once daily.

Key inclusion criteria:

Patients diagnosed with Parkinson’s disease according to the UK Parkinson’s Disease Society (UKPDS) brain bank diagnostic criteria and aged 30 years or older were eligible for the study if all of the following criteria were met.

- Modified Hoehn & Yahr stages 2-4 in the OFF state
- Responsive to levodopa/dopa-decarboxylase inhibitor (DCI) for at least 1 year with a stable regimen for at least 4 weeks before the baseline visit
- Taking at least 3 doses of levodopa/DCI per day with predictable end-of-dose wearing off
- An average of at least 120 minutes of OFF time per day on the two diaries during the 7-day period preceding the baseline visit

- On a stable PD regimen* within normal therapeutic limits for at least 4 weeks before the baseline visit

Levodopa/DCI and other anti-PD medications could not be altered (no changes in regimen or dose and no additional medications) until Week 16 (at the end of the follow-up period) or 4 weeks after treatment discontinuation, unless adverse events that were strongly suspected of being related to these medications occurred.

All of 89 randomized subjects (30 subjects in the placebo group, 31 subjects in the 20 mg group, 28 subjects in the 40 mg group) received study drug and at least one valid diary after the start of study treatment was collected from all subjects. Thus, 89 subjects were included in the safety analysis set and in the Full Analysis Set (FAS) and the FAS was used for the primary efficacy analysis. There were 10 withdrawals (2 subjects, 3 subjects, and 5 subjects, respectively) and the main reason for withdrawals was the occurrence of adverse events in 7 subjects (1 subject, 1 subject, and 5 subjects, respectively).

In the FAS, the mean daily levodopa doses at the baseline visit were 465.0 mg in the placebo group, 438.7 mg in the 20 mg group, and 450.0 mg in the 40 mg group.

The primary efficacy endpoint of the change from baseline to endpoint (Last Observation Carried Forward [LOCF]) in the mean percentage of awake time per day spent in the OFF state was as shown in Table 4. Concerning the secondary endpoint of the mean total hours of awake time per day spent in the OFF state, the baseline values (mean \pm SD) were 6.42 ± 2.52 hours in the placebo group, 6.08 ± 2.42 hours in the 20 mg group, and 6.45 ± 3.54 hours in the 40 mg group and the least-squares mean changes from baseline to endpoint (LOCF) [95% CI]¹ were -1.11 [-2.09, -0.14], -1.10 [-2.06, -0.13], and -1.50 [-2.51, -0.49] hours, respectively.

* levodopa/DCI, centrally acting dopamine agonists (bromocriptine, pergolide, cabergoline, talipexole, pramipexole), other anti-PD medications (anticholinergics, selegiline, droxidopa, amantadine, antihistamines, β -blockers)

¹ Calculated using an ANCOVA model with treatment as a factor and the baseline value as a covariate.

Table 4. Change in the mean percentage of awake time per day spent in the OFF state (%)

	Placebo (N = 30)	20 mg (N = 31)	40 mg (N = 28)
Baseline (Mean \pm SD)	39.65 \pm 15.32	39.04 \pm 16.58	39.07 \pm 16.90
Endpoint (LOCF) (Mean \pm SD)	31.42 \pm 21.74	34.20 \pm 18.70	31.41 \pm 23.23
Change (Least-squares mean [95% CI]) ^a	-8.10 [-14.72, -1.49]	-4.90 [-11.41, 1.61]	-7.72 [-14.57, -0.87]
Difference from placebo ^b (Least-squares mean [95% CI]) ^a	—	3.20 [-6.08, 12.48]	0.38 [-9.14, 9.91]
<i>P</i> -value ^a versus placebo	—	<i>P</i> = 0.495	<i>P</i> = 0.936

a: Calculated using an ANCOVA model with treatment as a factor and the baseline value as a covariate

b: istradefylline minus placebo

Concerning the secondary endpoint of the Unified Parkinson's disease rating scale (UPDRS) part III total score in the ON state, the scores at the baseline visit (Mean \pm SD) were 20.5 \pm 10.7 in the placebo group, 23.8 \pm 13.8 in the 20 mg group, and 22.7 \pm 12.9 in the 40 mg group and the least-squares mean changes from baseline to endpoint (LOCF) [95% CI]^{*} were -2.5 [-4.8, -0.2], -3.9 [-6.2, -1.7], and -5.0 [-7.3, -2.7], respectively.

Regarding safety, the incidence of adverse events was 60.0% (18 of 30 subjects) in the placebo group, 80.6% (25 of 31 subjects) in the 20 mg group, and 75.0% (21 of 28 subjects) in the 40 mg group. Adverse events reported by at least 5% of subjects in any group are shown in Table 5.

^{*} Calculated using an ANCOVA model with treatment as a factor and the baseline value as a covariate.

Table 5. Adverse events reported by at least 5% of subjects in any group

	Placebo (N = 30)	20 mg (N = 31)	40 mg (N = 28)
Overall incidence	60.0 (18)	80.6 (25)	75.0 (21)
Dyskinesia	3.3 (1)	16.1 (5)	17.9 (5)
Nausea	6.7 (2)	6.5 (2)	17.9 (5)
Nasopharyngitis	0 (0)	19.4 (6)	14.3 (4)
Constipation	3.3 (1)	6.5 (2)	10.7 (3)
Dizziness postural	6.7 (2)	3.2 (1)	10.7 (3)
Accident	3.3 (1)	3.2 (1)	10.7 (3)
Weight decreased	3.3 (1)	3.2 (1)	10.7 (3)
Urinary sediment abnormal	3.3 (1)	3.2 (1)	10.7 (3)
Hallucination	0 (0)	3.2 (1)	10.7 (3)
Blood trypsin increased	6.7 (2)	3.2 (1)	7.1 (2)
Decreased appetite	3.3 (1)	3.2 (1)	7.1 (2)
Contusion	3.3 (1)	0 (0)	7.1 (2)
Parkinson's disease	0 (0)	0 (0)	7.1 (2)
Headache	0 (0)	9.7 (3)	3.6 (1)
Blood CK increased	6.7 (2)	6.5 (2)	3.6 (1)
Lipase increased	6.7 (2)	3.2 (1)	3.6 (1)
Back pain	6.7 (2)	3.2 (1)	3.6 (1)
Blood LDH increased	6.7 (2)	0 (0)	3.6 (1)
Thirst	0 (0)	6.5 (2)	0 (0)
Blood urea increased	6.7 (2)	3.2 (1)	0 (0)
Hallucination, visual	6.7 (2)	3.2 (1)	0 (0)
γ -GT increased	10.0 (3)	0 (0)	0 (0)

% (N), LDH: lactate dehydrogenase, GT: glutamyl transferase

No deaths were reported. Serious adverse events were reported by 1 subject in the 20 mg group (pneumonia) and 2 subjects in the 40 mg group (hyperventilation and anxiety; hallucination and persecutory delusion) and a causal relationship to study drug could not be denied for hallucination and persecutory delusion in the 40 mg group. All of the serious adverse events resolved following the discontinuation of study drug or during continued study treatment.

Adverse events leading to treatment discontinuation were reported by 1 subject in the placebo group (aggression, visual hallucinations, delusion), 1 subject in the 20 mg group (dyskinesia), and 5 subjects in the 40 mg group (hallucination, anorexia, and persecutory delusion; nausea, dysgeusia, decreased appetite, and weight decreased; Parkinson's disease; dyskinesia; headache, dyskinesia, pyrexia, and pharyngeal erythema).

4.(iii).A.(3).2) Japanese early phase II study of istradefylline as monotherapy (Study 6002-0407, Attached document 5.3.5.4-2 [Reference data]; Studied period, November 2005 to October 2006)

A randomized, double-blind, crossover comparative study was conducted at 19 centers in Japan to evaluate the efficacy and safety of istradefylline as monotherapy in patients with Parkinson's disease (Target number of subjects, 32 subjects per sequence, a total of 64 subjects).

A 2-week screening period was followed by a 4-week double-blind treatment period (Period I and Period II) in which 40 mg of istradefylline or placebo was orally administered once daily in a crossover fashion (a 4-week washout period).

Key inclusion criteria were: diagnosed with Parkinson's disease according to the UKPDS brain bank diagnostic criteria; modified Hoehn & Yahr stages 1-3; not taking anti-PD medications*; and aged 30 years or older.

As all of 73 randomized subjects (37 subjects in Sequence A, 36 subjects in Sequence B) received study drug, 73 subjects were included in the safety analysis set. Of the randomized subjects, 72 subjects excluding 1 subject (Sequence B) (No on-treatment UPDRS part III scores were available in either Period I or Period II) were included in the FAS, which was used for the primary efficacy analysis. There were 8 withdrawals (2 subjects and 6 subjects, respectively) and the main reason for withdrawals was the occurrence of adverse events in 5 subjects (2 subjects and 3 subjects, respectively).

The primary efficacy endpoint of the change in the UPDRS part III total score from baseline to the end of each treatment period (LOCF) was as shown in Table 6.

* Patients who had taken levodopa/DCI, centrally acting dopamine agonists, anticholinergics, droxidopa, amantadine, or promethazine within 4 weeks before the baseline visit, patients who had taken selegiline within 3 months before the baseline visit, and patients who had previously been treated with levodopa/DCI for ≥ 4 weeks were excluded from the study.

Table 6. Change in the UPDRS part III total score

	Placebo (N = 70)	40 mg (N = 71)
Baseline (Mean \pm SD)	16.5 \pm 7.1	16.5 \pm 8.0
Endpoint (LOCF) (Mean \pm SD)	14.6 \pm 7.8	14.9 \pm 8.1
Change (Least-squares mean [95% CI]) ^a	-1.9 [-2.9, -0.8]	-1.6 [-2.6, -0.5]
Difference from placebo ^b (Least-squares mean [95% CI]) ^a	—	0.3 [-1.2, 1.7]
<i>P</i> -value ^a versus placebo	—	<i>P</i> = 0.699

a: Calculated using a mixed effect model with subject as a random effect and sequence, period, treatment, and baseline score of each period as fixed effects (N = 72)

b: Istradefylline minus placebo

Regarding safety, the incidence of adverse events was 39.4% (28 of 71 subjects) with placebo and 51.4% (36 of 70 subjects) with 40 mg istradefylline. Adverse events with an incidence of $\geq 5\%$ with either treatment were nasopharyngitis only [7.0% (5 of 71 subjects) with placebo, 5.7% (4 of 70 subjects) with 40 mg istradefylline].

No deaths were reported. Serious adverse events were reported by 1 subject after administration of placebo (Parkinsonism) and 1 subject after administration of 40 mg of istradefylline (congestive cardiomyopathy, ventricular tachycardia) and a causal relationship to study drug could not be denied for ventricular tachycardia occurring after administration of 40 mg of istradefylline and the event had an outcome of “unresolved.”

Adverse events leading to treatment discontinuation occurred in 2 subjects after administration of placebo (ALT increased, AST increased, and aldolase increased; nausea, insomnia, and feeling abnormal) and 3 subjects after administration of 40 mg of istradefylline (vestibular disorder, headache, and dizziness postural; convulsion; Parkinsonism).

4.(iii).A.(3).3 Japanese late phase II study (Study 6002-0608, Attached document 5.3.5.1-3; Studied period, March 2007 to August 2008)

A randomized, double-blind, parallel-group, comparative study was conducted at 47 centers in Japan to evaluate the efficacy and safety of istradefylline in levodopa-treated Parkinson's disease patients with motor complications (Target number of subjects, 120 subjects per group, a total of 360 subjects).

A 2-week screening period was followed by a 12-week double-blind treatment period in which 20 or 40 mg of istradefylline or placebo was orally administered once daily.

Key inclusion criteria:

Patients diagnosed with Parkinson's disease according to the UKPDS brain bank diagnostic criteria and aged 20 years or older were eligible for the study if all of the following criteria were met.

- Modified Hoehn & Yahr stages 2-4 in the OFF state at the baseline visit
- Responsive to levodopa/DCI for at least 1 year before the baseline visit
- Taking at least 3 doses and ≥ 300 mg of levodopa/DCI per day for at least 4 weeks before the baseline visit with predictable end-of-dose wearing off
- An average of at least 2 hours of OFF time per day on the four or more valid diaries during the 7-day period preceding the baseline visit
- On a stable PD regimen* (no additional medications and no changes in regimen) for at least 4 weeks before the baseline visit

Levodopa/DCI and other anti-PD medications could not be altered (no changes in regimen or dose and no additional medications) during the study period, unless adverse events that were strongly suspected of being related to these medications occurred.

Of 363 randomized subjects (119 subjects in the placebo group, 119 subjects in the 20 mg group, 125 subjects in the 40 mg group), 362 subjects excluding 1 subject in the 20 mg group received study drug. Thus, 362 subjects were included in the safety analysis set. Excluding 5 subjects without four or more valid diaries at all timepoints (1 subject, 3 subjects, and 1 subject, respectively) from the safety analysis set, 357 subjects were included in the FAS, which was used for the primary efficacy analysis. There were 36 withdrawals (10 subjects, 13 subjects, and 13 subjects, respectively) and the main reason for withdrawals was the occurrence of adverse events in 17 subjects (2 subjects, 7 subjects, and 8 subjects, respectively).

In the FAS, the mean daily levodopa doses at the baseline visit were 426.3 mg in the placebo group, 407.0 mg in the 20 mg group, and 415.3 mg in the 40 mg group.

The primary efficacy endpoint of the change in the mean total hours of awake time per day spent in the OFF state was as shown in Table 7.

* Drugs coded as 87116X according to the Standard Commodity Classification No. of Japan and promethazine hydrochloride

Table 7. Change in the mean total hours of awake time per day spent in the OFF state (hours)

	Placebo (N = 118)	20 mg (N = 115)	40 mg (N = 124)
Baseline (Mean \pm SD)	6.43 \pm 2.71	6.79 \pm 2.86	6.55 \pm 2.48
Endpoint (LOCF) (Mean \pm SD)	5.80 \pm 3.27	5.42 \pm 3.37	4.99 \pm 2.68
Change (Least-squares mean [95% CI]) ^a	-0.66 [-1.08, -0.25]	-1.31 [-1.73, -0.89]	-1.58 [-1.99, -1.17]
Difference from placebo ^b (Least-squares mean [95% CI]) ^a	—	-0.65 [-1.23, -0.07]	-0.92 [-1.49, -0.35]
<i>P</i> -value ^{a,c} versus placebo	—	<i>P</i> = 0.013	<i>P</i> < 0.001

a: Calculated using an ANCOVA model with treatment as a factor and the baseline value and center as covariates.

b: Istradefylline minus placebo, c: Williams test (one-sided level of significance of 0.025)

Concerning the secondary endpoint of the UPDRS part III total score in the ON state, the scores at the baseline visit (mean \pm SD) were 20.6 \pm 9.2 in the placebo group, 21.0 \pm 10.6 in the 20 mg group, and 21.1 \pm 11.0 in the 40 mg group and the least-squares mean changes from baseline to endpoint (LOCF) [95% CI]^{*} were -3.7 [-4.8, -2.6], -5.7 [-6.8, -4.6], and -5.7 [-6.8, -4.6], respectively.

Regarding safety, the incidence of adverse events was 58.0% (69 of 119 subjects) in the placebo group, 59.3% (70 of 118 subjects) in the 20 mg group, and 59.2% (74 of 125 subjects) in the 40 mg group. Adverse events reported by at least 5% of subjects in any group are shown in Table 8.

Table 8. Adverse events reported by at least 5% of subjects in any group

	Placebo (N = 119)	20 mg (N = 118)	40 mg (N = 125)
Overall incidence	58.0 (69)	59.3 (70)	59.2 (74)
Nasopharyngitis	4.2 (5)	5.9 (7)	8.8 (11)
Dyskinesia	2.5 (3)	8.5 (10)	6.4 (8)
Constipation	5.9 (7)	5.1 (6)	6.4 (8)
Protein urine present	2.5 (3)	1.7 (2)	6.4 (8)
Upper respiratory tract inflammation	0.8 (1)	3.4 (4)	5.6 (7)
Blood CK increased	3.4 (4)	6.8 (8)	4.8 (6)
Blood trypsin increased	5.0 (6)	1.7 (2)	4.0 (5)
Blood urine present	0.8 (1)	5.9 (7)	2.4 (3)

% (N)

No deaths were reported. Serious adverse events were reported by 2 subjects in the placebo group (skin tear and blood CK increased; transient ischaemic attack), 3 subjects in the 20 mg group (back pain and spinal compression fracture; contusion; gastric ulcer), and 6 subjects in the 40 mg group (emphysema; blood CK increased and neuropathy peripheral; cough; hypertension; cholecystitis; depression and persecutory

* Calculated using an ANCOVA model with treatment as a factor and the baseline value and center as covariates.

delusion) and a causal relationship to study drug could not be denied for transient ischaemic attack in the placebo group, gastric ulcer in the 20 mg group, and cough, hypertension, depression, and persecutory delusion in the 40 mg group. All of the serious adverse events improved or resolved following the discontinuation of study drug or during continued study treatment.

The incidence of adverse events leading to treatment discontinuation was 1.7% (2 of 119 subjects) in the placebo group, 5.9% (7 of 118 subjects) in the 20 mg group, and 6.4% (8 of 125 subjects) in the 40 mg group and those reported by at least 2 subjects in any group were dyskinesia (0 subjects, 3 subjects, and 1 subject, respectively), hallucination (0 subjects, 0 subjects, and 2 subjects, respectively), AST increased (0 subjects, 0 subjects, and 2 subjects, respectively), blood alkaline phosphatase increased (0 subjects, 0 subjects, and 2 subjects, respectively), and Parkinson's disease (0 subjects, 2 subjects, and 0 subjects, respectively).

4.(iii).A.(3).4 Japanese phase III study (Study 6002-009, Attached document 5.3.5.1-5; Studied period, July 2009 to February 2011)

A randomized, double-blind, parallel-group, comparative study was conducted at 44 centers in Japan to confirm the efficacy of istradefylline in levodopa-treated Parkinson's disease patients with motor complications (Target number of subjects, 120 subjects per group, a total of 360 subjects).

A 2- to 4-week screening period was followed by a 12-week double-blind treatment period in which 20 or 40 mg of istradefylline or placebo was orally administered once daily.

Key inclusion criteria and the rules for concomitant anti-PD medications were the same as those in the Japanese late phase II study.

As all of 373 randomized subjects (126 subjects in the placebo group, 123 subjects in the 20 mg group, 124 subjects in the 40 mg group) received study drug, 373 subjects were included in the safety analysis set. Excluding 7 subjects without four or more valid diaries at all timepoints (3 subjects, 3 subjects, and 1 subject, respectively) from the safety analysis set, 366 subjects were included in the FAS, which was used for the primary efficacy analysis. There were 38 withdrawals (17 subjects, 12 subjects, and 9 subjects, respectively) and the main reasons for withdrawals were adverse events in 17 subjects (6 subjects, 5 subjects, and 6 subjects, respectively) and subject's request in 15 subjects (6 subjects, 6 subjects, and 3 subjects, respectively).

In the FAS, the mean daily levodopa doses at the baseline visit were 425.4 mg in the placebo group, 430.8 mg in the 20 mg group, and 420.5 mg in the 40 mg group.

The primary efficacy endpoint of the change in the mean total hours of awake time per day spent in the OFF state was as shown in Table 9.

Table 9. Change in the mean total hours of awake time per day spent in the OFF state (hours)

	Placebo (N = 123)	20 mg (N = 120)	40 mg (N = 123)
Baseline (Mean ± SD)	6.31 ± 2.47	6.55 ± 2.72	5.97 ± 2.45
Endpoint (LOCF) (Mean ± SD)	6.03 ± 3.05	5.46 ± 2.93	5.00 ± 2.84
Change (Least-squares mean [95% CI]) ^a	-0.23 [-0.62, 0.16]	-0.99 [-1.38, -0.60]	-0.96 [-1.35, -0.58]
Difference from placebo ^b (Least-squares mean [95% CI]) ^a	—	-0.76 [-1.30, -0.22]	-0.74 [-1.27, -0.20]
<i>P</i> -value ^{a,c} versus placebo	—	<i>P</i> = 0.003	<i>P</i> = 0.003

a: Calculated using an ANCOVA model with treatment as a factor and the baseline value and center as covariates.

b: Istradefylline minus placebo, c: Williams test (one-sided level of significance of 0.025)

Concerning the secondary endpoint of the UPDRS part III total score in the ON state, the scores at the baseline visit (Mean ± SD) were 21.6 ± 11.6 in the placebo group, 21.3 ± 10.8 in the 20 mg group, and 20.7 ± 11.0 in the 40 mg group and the least-squares mean changes from baseline to endpoint (LOCF) [95% CI]^{*} were -2.8 [-3.8, -1.8], -3.7 [-4.7, -2.8], and -4.9 [-5.8, -3.9], respectively.

Regarding safety, the incidence of adverse events was 51.6% (65 of 126 subjects) in the placebo group, 65.0% (80 of 123 subjects) in the 20 mg group, and 59.7% (74 of 124 subjects) in the 40 mg group. Adverse events reported by at least 5% of subjects in any group are shown in Table 10.

Table 10. Adverse events reported by at least 5% of subjects in any group

	Placebo (N = 126)	20 mg (N = 123)	40 mg (N = 124)
Overall incidence	51.6 (65)	65.0 (80)	59.7 (74)
Dyskinesia	4.0 (5)	13.0 (16)	12.1 (15)
Nasopharyngitis	8.7 (11)	8.1 (10)	5.6 (7)
Constipation	2.4 (3)	5.7 (7)	3.2 (4)
Somnolence	3.2 (4)	6.5 (8)	1.6 (2)
Blood CK increased	5.6 (7)	3.3 (4)	0.8 (1)

% (N)

One death occurred in the placebo group (death) and as the subject was found dead and the details of his death were unknown, its causal relationship to study drug could not be denied. Serious adverse events were reported by 2 subjects in the placebo group (toxicity to various agents; breast cancer in situ), 6 subjects in the 20 mg group (pneumonia bacterial; gait disturbance; radius fracture; sciatica and neuralgia; Parkinsonism and delirium; bile duct cancer), and 6 subjects in the 40 mg group (gastric ulcer; bronchitis; myocardial infarction; pneumonia aspiration; hallucination; rectal cancer) and a causal relationship to study drug could not be denied for gait disturbance and Parkinsonism in the 20 mg group and gastric ulcer, myocardial

^{*} Calculated using an ANCOVA model with treatment as a factor and the baseline value and center as covariates

infarction, and hallucination in the 40 mg group. All of the serious adverse events excluding bile duct cancer in the 20 mg group improved or resolved following the discontinuation of study drug or during continued study treatment.

The incidence of adverse events leading to treatment discontinuation was 4.0% (5 of 126 subjects) in the placebo group, 4.1% (5 of 123 subjects) in the 20 mg group, and 4.8% (6 of 124 subjects) in the 40 mg group and those reported by at least 2 subjects in any group were dyskinesia (0 subjects, 0 subjects, and 2 subjects, respectively).

4.(iii).A.(3).5) Japanese long-term treatment study (Study 6002-010, Attached document 5.3.5.2-1; Studied period, October 2009 to March 2012)

An open-label study was conducted in subjects who completed a Japanese phase III study (Study 6002-009) at 44 centers in Japan to evaluate the safety and efficacy of long-term treatment with istradefylline (Target number of subjects of ≥ 150).

In the 1- to 4-week transition phase of the study, subjects orally received their dose of study drug assigned in Study 6002-009 (istradefylline 20 mg, istradefyllineb 40 mg, or placebo) once daily and those eligible for participation in the long-term treatment study were immediately entered into the open-label phase of the study. The first 8 weeks of the open-label phase were the dose titration period and istradefylline was initiated at a dose of 20 mg orally once daily. If all of the criteria (no safety problems, inadequate response, subject's request for a dose increase) were met and the investigator etc. considered it appropriate to increase the dose at Week 4 of the open-label phase, the dose was to be increased to 40 mg. After the dose was increased to 40 mg, if either one of the two criteria (safety problems [the occurrence of adverse events etc.], excessive response) was met and the investigator etc. considered it appropriate to decrease the dose at Week 8 of the open-label phase, the dose was to be decreased to 20 mg. During the maintenance period (from Week 8 to Week 52 of the open-label phase), as a rule, no changes in dose were permitted. Even during the maintenance period, if all of the dose increase criteria, like those for the dose titration period, were met, the dose was to be increased to 40 mg. For subjects treated with 40 mg in the maintenance period, if there was a safety problem and the investigator etc. considered it appropriate to decrease the dose, the dose was to be decreased to 20 mg. However, once the dose was decreased from 40 mg to 20 mg during the maintenance period, increasing the dose from 20 mg to 40 mg again was prohibited.

Key inclusion criteria were patients who completed the double-blind treatment period (12 weeks) of Study 6002-009. Anti-PD medications could not be altered (no changes in regimen or dose and no additional medications) during the transition phase and the dose titration period, unless adverse events that were strongly suspected of being related to these medications occurred. Although anti-PD medications that subjects were taking were not to be altered (no changes in regimen or dose and no additional medications) wherever possible during the maintenance period, dose increases of anti-PD medications and the use of additional medications were allowed in subjects who responded inadequately to an increased dose of 40 mg

of istradefylline or subjects treated with 20 mg of istradefylline for safety reasons who responded inadequately.

Of the subjects who completed Study 6002-009, 313 subjects (103 subjects in the placebo group, 102 subjects in the 20 mg group, 108 subjects in the 40 mg group) entered the transition phase and 308 subjects (100 subjects, 101 subjects, and 107 subjects, respectively) excluding 5 subjects withdrawn during the transition phase (subject's request [3 subjects], exclusion criteria violations [2 subjects]) entered the open-label phase and all of the subjects who entered the open-label phase were included in the safety analysis set. Excluding 1 subject without four or more valid diaries at all timepoints during the open-label phase (1 subject, 0 subjects, and 0 subjects, respectively) from the safety analysis set, 307 subjects were included in the FAS, which was used for the primary efficacy analysis. There were 77 withdrawals (27 subjects, 28 subjects, and 22 subjects, respectively) during the open-label phase and the main reasons for withdrawals were subject's request in 29 subjects (10 subjects, 13 subjects, and 6 subjects, respectively) and adverse events in 24 subjects (13 subjects, 6 subjects, and 5 subjects, respectively). Among 296 subjects who were treated with 20 mg of istradefylline at Week 4 of the open-label phase and continued to receive istradefylline thereafter, 132 subjects were maintained on 20 mg of istradefylline and 164 subjects had their dose increased to 40 mg.

In the FAS, the mean daily levodopa doses at the baseline visit in Study 6002-009 were 415.9 mg in the placebo group, 424.3 mg in the 20 mg group, and 422.7 mg in the 40 mg group.

Regarding efficacy, the mean total hours of awake time per day spent in the OFF state by treatment group in Study 6002-009 was as shown in Figure 1. The baseline values (the values obtained during the transition phase) (mean \pm SD) were 5.98 ± 3.14 hours in the placebo group, 5.39 ± 2.98 hours in the 20 mg group, and 4.91 ± 2.71 hours in the 40 mg group and the changes from baseline to Week 52 were -0.40 ± 2.27 , -0.59 ± 2.71 , and 0.26 ± 2.71 hours, respectively.

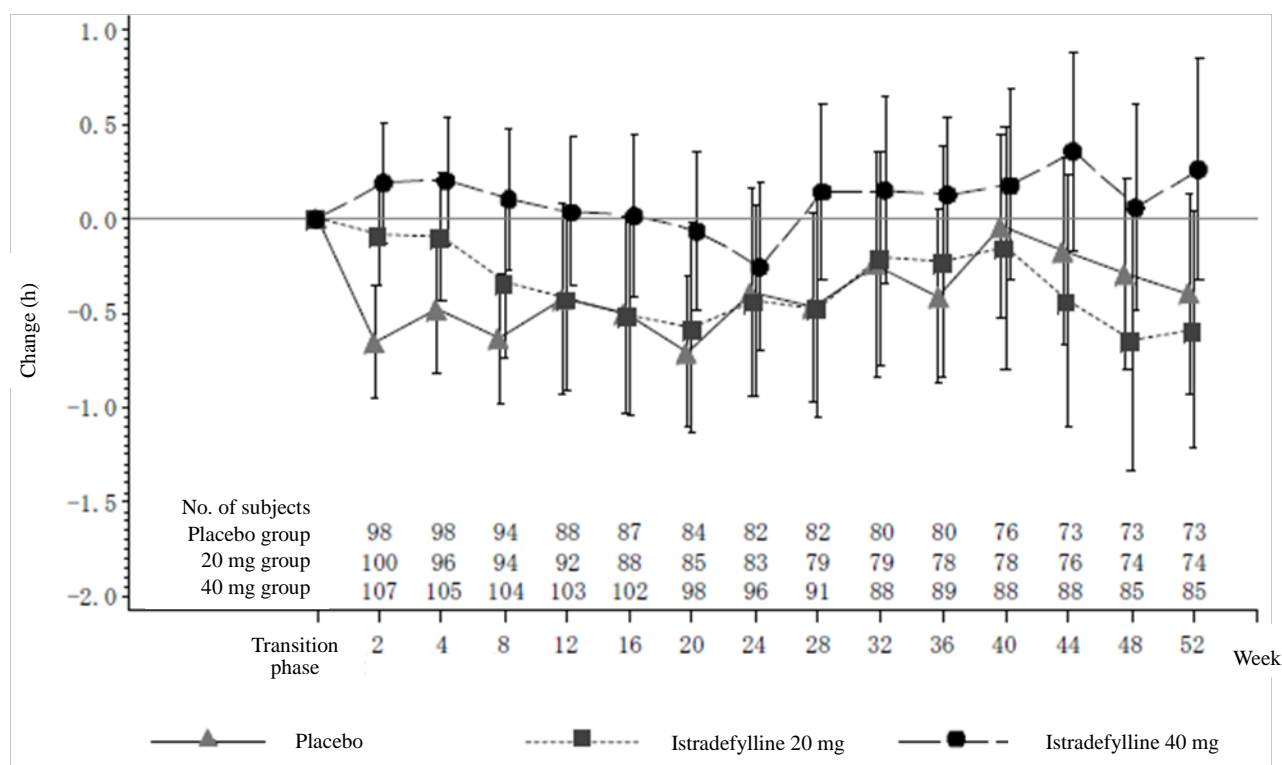


Figure 1. Change in the mean total hours of awake time per day spent in the OFF state (Mean and 95% CI)
(Adapted from Attached document 5.3.5.2-1 Figure 11.4.1.1-1)

The UPDRS part III total scores in the ON state at baseline were 18.1 ± 11.5 in the placebo group, 17.8 ± 10.8 in the 20 mg group, and 16.1 ± 10.8 in the 40 mg group and the changes from baseline to Week 52 were -3.4 ± 6.1 , -1.7 ± 7.6 , and -1.6 ± 6.0 , respectively.

Regarding safety, the incidence of adverse events was 88.0% (271 of 308 subjects) and adverse events with an incidence of $\geq 5\%$ are shown in Table 11.

Table 11. Adverse events with an incidence of $\geq 5\%$

	N = 308
Overall incidence	88.0 (271)
Nasopharyngitis	24.4 (75)
Dyskinesia	21.4 (66)
Contusion	10.4 (32)
Constipation	9.4 (29)
Hallucination, visual	8.8 (27)
Weight decreased	7.1 (22)
Insomnia	5.2 (16)

% (N)

No deaths occurred. Serious adverse events occurred in 43 subjects during the transition phase or open-label phase (4 subject during the transition phase, 39 subjects during the open-label phase) and those reported by

at least 2 subjects were spinal compression fracture (4 subjects), colonic polyp (2 subjects), femoral neck fracture (2 subjects), cataract (2 subjects), and gastric ulcer haemorrhage (2 subjects) and a causal relationship to study drug could not be denied for 1 case of gastric ulcer haemorrhage, but the event had an outcome of “resolved/recovered.”

In the transition phase and the open-label phase, the incidence of adverse events leading to treatment discontinuation was 8.0% (25 of 313 subjects) (1 subject during the transition phase, 24 subjects during the open-label phase) and those reported by at least 2 subjects were hallucination (4 subjects), visual hallucinations (2 subjects), dyskinesia (2 subjects), and delusion (2 subjects).

4.(iii).A.(4) Foreign clinical studies in patients with Parkinson’s disease

4.(iii).A.(4).1) US phase III study (a) (Study 6002-US-013, Attached document 5.3.5.1-9 [Reference data]; Studied period, July 2004 to November 2005)

A randomized, double-blind, parallel-group, comparative study was conducted at 26 centers overseas to evaluate the efficacy and safety of istradefylline in levodopa/carbidopa-treated Parkinson’s disease patients with motor complications (Target number of subjects, 105 subjects per group, a total of 210 subjects).

A 2- to 8-week screening period was followed by a 12-week double-blind treatment period in which 20 mg of istradefylline or placebo was orally administered once daily.

Key inclusion criteria:

Patients diagnosed with Parkinson’s disease according to the UKPDS brain bank diagnostic criteria and aged 30 years or older were eligible for the study if all of the following criteria were met.

- Modified Hoehn & Yahr stages 2-4 in the OFF state
- Responsive to levodopa/carbidopa for at least 1 year before randomization
- Taking at least 3 doses of levodopa per day with predictable end-of-dose wearing off
- An average of at least 180 minutes of OFF time per day on the two valid diaries prior to the baseline visit

Levodopa and other anti-PD medications could not be altered (no changes in regimen or dose and no additional medications) during the study period, unless adverse events that were strongly suspected of being related to these medications occurred.

Of 231 randomized subjects (115 subjects in the placebo group, 116 subjects in the 20 mg group), 230 subjects excluding 1 subject in the 20 mg group received study drug. Thus, 230 subjects were included in the safety analysis set and 225 subjects with diary data at baseline (before the start of study treatment) and after receiving at least one dose of study drug (113 subjects and 112 subjects, respectively) were included in the Intent-to-Treat (ITT) population, which was used for the primary efficacy analysis. There were 24 withdrawals (14 subjects and 14 subjects, respectively) and the main reasons for withdrawals were adverse

events in 13 subjects (7 subjects and 6 subjects, respectively) and deviations in 6 subjects (1 subject and 5 subjects, respectively).

In the safety analysis set, the mean daily levodopa doses at baseline were 630.7 mg in the placebo group and 652.4 mg in the 20 mg group.

The primary efficacy endpoint of the change from baseline to endpoint (LOCF) in the mean percentage of awake time per day spent in the OFF state was as shown in Table 12.

Table 12. Change in the mean percentage of awake time per day spent in the OFF state (%)

	Placebo (N = 113)	20 mg (N = 112)
Baseline (Mean \pm SD)	38.72 \pm 11.612	39.81 \pm 14.029
Endpoint (LOCF) (Mean \pm SD)	33.74 \pm 17.416	30.48 \pm 16.315
Change (Least-squares mean [95% CI]) ^a	-4.92 [-7.76, -2.08]	-9.49 [-12.43, -6.56]
Difference from placebo ^b (Least-squares mean [95% CI]) ^a	—	-4.57 [-8.55, -0.59]
<i>P</i> -value ^a versus placebo	—	<i>P</i> = 0.025

a: Calculated using an ANCOVA model with investigator and treatment as factors and the baseline value as a covariate.

b: Istradefylline minus placebo

Concerning the secondary endpoint of the UPDRS part III total score in the ON state, the baseline scores (mean \pm SD) were 22.8 \pm 11.13 in the placebo group and 24.0 \pm 11.34 in the 20 mg group and the least-squares mean changes from baseline to endpoint [95% CI]^{*} were -2.0 [-3.5, -0.4] and -2.9 [-4.5, -1.3], respectively.

Regarding safety, the incidence of adverse events was 75.7% (87 of 115 subjects) in the placebo group and 79.1% (91 of 115 subjects) in the 20 mg group. Adverse events reported by at least 5% of subjects in either group are shown in Table 13.

^{*}Calculated using an ANCOVA model with investigator and treatment as factors and the baseline value as a covariate.

Table 13. Adverse events reported by at least 5% of subjects in either group

	Placebo (N = 115)	20 mg (N = 115)
Overall incidence	75.7 (87)	79.1 (91)
Dyskinesia	12.2 (14)	22.6 (26)
Nausea	7.0 (8)	7.8 (9)
Weight increased	7.0 (8)	7.8 (9)
Light headedness	3.5 (4)	7.8 (9)
Parkinson's disease	6.1 (7)	7.0 (8)
Arthralgia	7.0 (8)	6.1 (7)
Weight decreased	2.6 (3)	6.1 (7)
Tremor	2.6 (3)	5.2 (6)
Constipation	0.9 (1)	5.2 (6)
Accident	7.0 (8)	4.3 (5)
Insomnia	5.2 (6)	0.9 (1)

% (N)

One death occurred in the placebo group (sudden death) and its causal relationship to study drug could not be denied. Serious adverse events were reported by 4 subjects in the placebo group (cor pulmonale; pneumonia; cardiac disorder; coronary artery disease and haematoma) and 4 subjects in the 20 mg group (asthma, infection, urosepsis, cardiac failure) and a causal relationship to study drug could not be denied for cor pulmonale and cardiac disorder in the placebo group and asthma and cardiac failure in the 20 mg group. All of the serious adverse events excluding cor pulmonale in the placebo group improved or resolved following the discontinuation of study drug or during continued study treatment.

The incidence of adverse events leading to treatment discontinuation was 6.1% (7 of 115 subjects) in the placebo group and 5.2% (6 of 115 subjects) in the 20 mg group and no individual events were reported by two or more subjects in either group.

4.(iii).A.(4).2) US phase III study (b) (Study 6002-US-018, Attached document 5.3.5.1-10 [Reference data]; Studied period, July 2004 to November 2005)

A randomized, double-blind, parallel-group, comparative study was conducted at 74 centers overseas to evaluate the efficacy and safety of istradefylline in levodopa/carbidopa-treated Parkinson's disease patients with motor complications (Target number of subjects, 140 subjects per group, a total of 560 subjects).

A 2- to 8-week screening period was followed by a 12-week double-blind treatment period in which 10, 20, or 40 mg of istradefylline or placebo was orally administered once daily.

Key inclusion criteria:

Patients diagnosed with Parkinson's disease according to the UKPDS brain bank diagnostic criteria and aged 30 years or older were eligible for the study if all of the following criteria were met.

- Modified Hoehn & Yahr stages 2-4 in the OFF state
- On levodopa preparations for at least 1 year and on a stable PD regimen including levodopa for at least 4 weeks before baseline
- Taking at least 3 doses of levodopa per day with predictable end-of-dose wearing off
- An average of at least 180 minutes of OFF time per day on the two diaries prior to the baseline visit

Levodopa and other anti-PD medications could not be altered (no changes in regimen or dose and no additional medications) during the study period, unless adverse events that were strongly suspected of being related to these medications occurred.

Of 610 randomized subjects (154 subjects in the placebo group, 155 subjects in the 10 mg group, 149 subjects in the 20 mg group, 152 subjects in the 40 mg group), 605 subjects excluding 5 subjects (3 subjects, 2 subjects, 0 subjects, and 0 subjects, respectively) received study drug. Thus, 605 subjects were included in the safety analysis set. The ITT population included 584 subjects with diary data at baseline (before the start of study treatment) and after receiving at least one dose of study drug (146 subjects, 149 subjects, 144 subjects, and 145 subjects, respectively), which was used for the primary efficacy analysis. There were 68 withdrawals (14 subjects, 19 subjects, 18 subjects, and 17 subjects, respectively) and the main reasons for withdrawals were adverse events in 43 subjects (7 subjects, 6 subjects, 15 subjects, and 15 subjects, respectively) and consent withdrawal in 10 subjects (3 subjects, 5 subjects, 1 subject, and 1 subject, respectively).

In the safety analysis set, the mean daily levodopa doses at baseline were 652.4 mg in the placebo group, 697.1 mg in the 10 mg group, 577.2 mg in the 20 mg group, and 623.7 mg in the 40 mg group.

The primary efficacy endpoint of the change from baseline to endpoint (LOCF) in the mean percentage of awake time per day spent in the OFF state was as shown in Table 14.

Table 14. Change in the mean percentage of awake time per day spent in the OFF state (%)

	Placebo (N = 146)	10 mg (N = 149)	20 mg (N = 144)	40 mg (N = 145)
Baseline (Mean \pm SD)	39.76 \pm 11.484	39.50 \pm 11.714	39.66 \pm 12.461	41.78 \pm 13.127
Endpoint (LOCF) (Mean \pm SD)	32.18 \pm 15.483	33.84 \pm 16.999	33.55 \pm 18.692	32.70 \pm 18.101
Change (Least-squares mean [95% CI]) ^a	-8.31 [-10.85, -5.78]	-6.52 [-9.03, -4.01]	-6.81 [-9.37, -4.26]	-8.97 [-11.51, -6.44]
Difference from placebo ^b (Least-squares mean [95% CI]) ^a	—	1.79 [-1.73, 5.31]	1.50 [-2.05, 5.05]	-0.66 [-4.21, 2.88]
<i>P</i> -value ^{a,c} versus placebo	—	<i>P</i> = 0.319	<i>P</i> = 0.408	<i>P</i> = 0.714

a: Calculated using an ANCOVA model with investigator and treatment as factors and the baseline value as a covariate.

b: Istradefylline minus placebo, c: closed testing procedure (Since there was no significant difference either between 40 mg and placebo or between 20 mg and placebo, the closed testing procedure was stopped. The *P*-value for the comparison of 10 mg against placebo is given as a reference.)

Concerning the secondary endpoint of the UPDRS part III total score in the ON state, the baseline scores (mean \pm SD) were 22.6 \pm 11.74 in the placebo group, 21.3 \pm 11.19 in the 10 mg group, 22.4 \pm 11.18 in the 20 mg group, and 22.0 \pm 11.44 in the 40 mg group and the least-squares mean changes from baseline to endpoint [95% CI]^{*} were -0.7 [-2.0, 0.6], -1.2 [-2.5, 0.1], -0.7 [-2.1, 0.6], and -2.8 [-4.1, -1.4], respectively.

Regarding safety, the incidence of adverse events was 76.2% (115 of 151 subjects) in the placebo group, 82.4% (126 of 153 subjects) in the 10 mg group, 83.9% (125 of 149 subjects) in the 20 mg group, and 84.2% (128 of 152 subjects) in the 40 mg group. Adverse events reported by at least 5% of subjects in any group are shown in Table 15.

^{*} Calculated using an ANCOVA model with investigator and treatment as factors and the baseline value as a covariate.

Table 15. Adverse events reported by at least 5% of subjects in any group

	Placebo (N = 151)	10 mg (N = 153)	20 mg (N = 149)	40 mg (N = 152)
Overall incidence	76.2 (115)	82.4 (126)	83.9 (125)	84.2 (128)
Dyskinesia	19.2 (29)	21.6 (33)	16.8 (25)	26.3 (40)
Insomnia	7.3 (11)	6.5 (10)	7.4 (11)	10.5 (16)
Nausea	6.0 (9)	7.2 (11)	4.7 (7)	7.9 (12)
Constipation	4.0 (6)	6.5 (10)	7.4 (11)	7.2 (11)
Light headedness	3.3 (5)	5.2 (8)	4.0 (6)	7.2 (11)
Headache	4.6 (7)	4.6 (7)	4.0 (6)	7.2 (11)
Accident	6.0 (9)	5.9 (9)	6.0 (9)	6.6 (10)
Arthralgia	6.0 (9)	7.8 (12)	2.7 (4)	5.9 (9)
Weight decreased	1.3 (2)	3.3 (5)	2.7 (4)	5.9 (9)
Hallucination	1.3 (2)	1.3 (2)	2.0 (3)	5.9 (9)
Upper respiratory tract infection	3.3 (5)	2.6 (4)	2.0 (3)	5.3 (8)
Parkinson's disease	5.3 (8)	11.1 (17)	6.0 (9)	4.6 (7)
Fatigue	4.6 (7)	3.9 (6)	6.7 (10)	3.9 (6)
Tremor	5.3 (8)	3.9 (6)	2.0 (3)	0.7 (1)

% (N)

One death each occurred in the placebo (cardio-respiratory arrest, mesenteric occlusion, intestinal gangrene) and 10 mg (cardiogenic shock) groups, but a causal relationship to study drug was denied for both cases. The incidence of serious adverse events was 4.0% (6 of 151 subjects) in the placebo group, 3.9% (6 of 153 subjects) in the 10 mg group, 6.0% (9 of 149 subjects) in the 20 mg group, and 5.3% (8 of 152 subjects) in the 40 mg group and those reported by at least 2 subjects in any group were accident (0 subjects, 1 subject, 0 subjects, and 2 subjects, respectively) and urinary tract infection (0 subjects, 0 subjects, 2 subjects, and 0 subjects, respectively) and a causal relationship to study drug could not be denied for accident in the 10 mg group and the event had an outcome of “sequelae.”

The incidence of adverse events leading to treatment discontinuation was 4.0% (6 of 151 subjects) in the placebo group, 3.9% (6 of 153 subjects) in the 10 mg group, 10.1% (15 of 149 subjects) in the 20 mg group, and 9.2% (14 of 152 subjects) in the 40 mg group and those reported by at least 2 subjects in any group were Parkinson's disease (2 subjects, 2 subjects, 1 subject, and 1 subject, respectively) and dyskinesia (1 subject, 0 subjects, 0 subjects, and 3 subjects, respectively).

4.(iii).A.(4).3 EU phase III study (Study 6002-EU-007, Attached document 5.3.5.1-11 [Reference data]; Studied period, November 2004 to October 2005)

A randomized, double-blind, parallel-group, comparative study was conducted at 59 centers overseas to evaluate the efficacy and safety of istradefylline or entacapone in levodopa-treated Parkinson's disease patients with motor complications (Target number of subjects, 135 subjects per group, a total of 405 subjects).

A 2-week screening period was followed by a 16-week double-blind treatment period in which 40 mg of istradefylline, 200 mg of entacapone, or placebo was orally administered. In the istradefylline group, 40 mg of istradefylline was administered with the first dose of the day of levodopa and placebo was administered with subsequent doses of levodopa.

Key inclusion criteria:

Patients diagnosed with Parkinson's disease according to the UKPDS brain bank diagnostic criteria and aged 30 years or older were eligible for the study if all of the following criteria were met.

- Modified Hoehn & Yahr stages 2-4 in the OFF state
- On levodopa preparations for at least 1 year and on a stable PD regimen including levodopa for at least 4 weeks before baseline
- Taking at least 3 doses of levodopa per day with predictable end-of-dose wearing off
- An average of at least 180 minutes of OFF time per day on the two diaries prior to the baseline visit, and not more than four invalid entries per diary.

Dose titration of levodopa was allowed during the first 4 weeks of the double-blind treatment period. Dose titration and discontinuation of other anti-PD medications and the use of additional medications were allowed during the first 4 weeks of the double-blind treatment period if symptoms were not controlled by levodopa or adverse events related to other anti-PD medications occurred.

As all of 464 randomized subjects (152 subjects in the placebo group, 159 subjects in the istradefylline 40 mg group, 153 subjects in the entacapone group) received study drug, 464 subjects were included in the safety analysis set and 455 subjects with diary data at baseline (before the start of study treatment) and after receiving at least one dose of study drug (151 subjects, 158 subjects, and 146 subjects, respectively) were included in the ITT population, which was used for the primary efficacy analysis. There were 54 withdrawals (19 subjects, 12 subjects, and 23 subjects, respectively) and the main reasons for withdrawals were adverse events in 27 subjects (10 subjects, 7 subjects, and 10 subjects, respectively) and consent withdrawal in 11 subjects (3 subjects, 2 subjects, and 6 subjects, respectively).

In the safety analysis set, the mean daily levodopa doses at baseline were 649.9 mg in the placebo group, 608.0 mg in the istradefylline 40 mg group, and 635.4 mg in the entacapone group.

The primary efficacy endpoint of the change from baseline to endpoint (LOCF) in the mean percentage of awake time per day spent in the OFF state was as shown in Table 16.

Table 16. Change in the mean percentage of awake time per day spent in the OFF state (%)

	Placebo (N = 151)	Istradefylline 40 mg (N = 158)	Entacapone (N = 146)
Baseline (Mean \pm SD)	41.49 \pm 12.257	38.61 \pm 11.625	40.05 \pm 13.423
Endpoint (LOCF) (Mean \pm SD)	36.73 \pm 17.311	34.30 \pm 16.095	32.63 \pm 17.741
Change (Least-squares mean [95% CI]) ^a	-4.53 [-7.02, 2.04]	-5.14 [-7.58, -2.69]	-7.82 [-10.34, -5.30]
Difference from placebo ^b (Least-squares mean [95% CI]) ^a	—	-0.61 [-4.05, 2.83]	-3.29 [-6.77, 0.19]
<i>P</i> -value ^a versus placebo	—	<i>P</i> = 0.729	<i>P</i> = 0.064

a: Calculated using an ANCOVA model with investigator and treatment as factors and the baseline value as a covariate

b: Istradefylline minus placebo

Concerning the secondary endpoint of the UPDRS part III total score in the ON state, the baseline scores (mean \pm SD) were 27.5 \pm 12.41 in the placebo group, 27.4 \pm 11.35 in the istradefylline 40 mg group, and 27.7 \pm 11.83 in the entacapone group and the least-squares mean changes from baseline to endpoint (LOCF) [95% CI]^{*} were -3.0 [-4.2, -1.8], -4.6 [-5.8, -3.4], and -4.8 [-6.0, -3.5], respectively.

Regarding safety, the incidence of adverse events was 63.8% (97 of 152 subjects) in the placebo group, 64.8% (103 of 159 subjects) in the istradefylline 40 mg group, and 66.0% (101 of 153 subjects) in the entacapone group. Adverse events reported by at least 5% of subjects in any group are shown in Table 17.

Table 17. Adverse events reported by at least 5% of subjects in any group

	Placebo (N = 152)	Istradefylline 40 mg (N = 159)	Entacapone (N = 153)
Overall incidence	63.8 (97)	64.8 (103)	66.0 (101)
Dyskinesia	7.2 (11)	13.8 (22)	13.1 (20)
Tremor	4.6 (7)	6.3 (10)	3.3 (5)
Back pain	2.6 (4)	6.3 (10)	2.6 (4)
Parkinson's disease	11.2 (17)	5.7 (9)	7.2 (11)
Headache	2.0 (3)	5.7 (9)	3.9 (6)
Constipation	4.6 (7)	5.0 (8)	7.8 (12)
Arthralgia	2.6 (4)	5.0 (8)	5.2 (8)
Nausea	4.6 (7)	3.8 (6)	8.5 (13)
Insomnia	3.9 (6)	3.8 (6)	5.9 (9)
Weight decreased	5.3 (8)	3.1 (5)	9.8 (15)
Light headedness	5.3 (8)	2.5 (4)	3.3 (5)

% (N)

One death in the placebo group (pneumonia and muscle rigidity) and two deaths in the entacapone group (acute respiratory distress syndrome; heat stroke) were reported, but a causal relationship to study drug was

^{*} Calculated using an ANCOVA model with investigator and treatment as factors and the baseline value as a covariate.

denied for all cases. The incidence of serious adverse events was 3.3% (5 of 152 subjects) in the placebo group, 3.1% (5 of 159 subjects) in the istradefylline 40 mg group, and 2.6% (4 of 153 subjects) in the entacapone group and no individual events were reported by two or more subjects in any group.

The incidence of adverse events leading to treatment discontinuation was 6.6% (10 of 152 subjects) in the placebo group, 4.4% (7 of 159 subjects) in the istradefylline 40 mg group, and 6.5% (10 of 153 subjects) in the entacapone group and those reported by at least 2 subjects in any group were dyskinesia (1 subject, 1 subject, and 2 subjects, respectively) and Parkinson's disease (3 subjects, 0 subjects, and 0 subjects, respectively).

4.(iii).B Outline of the review by PMDA

4.(iii).B.(1) Clinical positioning of istradefylline

PMDA asked the applicant to explain the clinical positioning of istradefylline in the treatment of Parkinson's disease, including the intended patient population, the expected usefulness of istradefylline, the choice between istradefylline and currently available anti-PD medications, and the need for their concomitant use.

The applicant responded as follows:

Japanese late phase II (Study 6002-0608) and Japanese phase III (Study 6002-009) studies included patients taking at least 3 doses and ≥ 300 mg of levodopa/DCI per day to ensure that among patients with modified Hoehn & Yahr stage 2 to 4 Parkinson's disease and at least 2 hours of OFF time per day, those experiencing wearing-off phenomena (ON/OFF fluctuations during the day) were selected. In the clinical studies, the efficacy of istradefylline in reducing the mean total hours of awake time per day spent in the OFF state (the primary endpoint) was confirmed and patients treated with istradefylline demonstrated an increase in ON time without troublesome dyskinesia (the secondary endpoint). Thus, the intended population for istradefylline is patients with Parkinson's disease on levodopa-containing preparations with motor complications and the high clinical usefulness of istradefylline in these patients, i.e. a reduction in OFF time and an increase in the ON time that is beneficial for patients, can be expected. Pooled analysis from Japanese placebo-controlled comparative studies (Study 6002-0406, Study 6002-0608, Study 6002-009) was performed to examine the impact of concomitant anti-PD medications on the change in the mean total hours of awake time per day spent in the OFF state. As a result, in all of the subgroups defined by concomitant medication, patients treated with istradefylline demonstrated a greater reduction in the mean total hours of awake time per day spent in the OFF state than those treated with placebo, indicating that istradefylline exerts its effects, regardless of the type or combination of other anti-PD medications used. On the other hand, regarding safety, the main adverse events reported in 4 Japanese studies in patients with Parkinson's disease (Study 6002-0406, Study 6002-0608, Study 6002-009, Study 6002-010) were dyskinesia, constipation, visual hallucinations, weight decreased, and nausea etc. Most of these adverse events were mild or moderate in severity and no clinically relevant adverse events occurred following administration of istradefylline, regardless of the combination or type of anti-PD medications used.

Based on the above, istradefylline with a novel mechanism of action involving adenosine A_{2A} receptor antagonism can be used efficaciously and safely in Parkinson's disease patients with motor complications, regardless of concomitant anti-PD medications and should be useful as a new pharmacotherapeutic option for Parkinson's disease. In addition, as once-daily administration of istradefylline is recommended and it is unnecessary to administer istradefylline simultaneously with a levodopa-containing preparation and no frequent dose adjustment is required, good compliance is also expected.

Concerning the choice between istradefylline and currently available anti-PD medications, according to the treatment algorithms recommended by "Parkinson's Disease Treatment Guideline 2011" of the Japanese Society of Neurology, for the treatment of wearing-off, if no improvement is noted despite optimized dopaminergic treatment with levodopa-containing preparations or dopamine agonists, treatment with entacapone, selegiline, or zonisamide should be initiated. The characteristics and problems of these anti-PD medications are described below.

Entacapone, like istradefylline, has been shown to improve the wearing-off phenomenon, but its effect in improving motor function in the ON state (improvement in the UPDRS part III score) has not been demonstrated clearly. With increasing frequency of administration per day, L-DOPA peaks (peak dose) are elevated towards evening, resulting in an increased risk of adverse drug reactions of dyskinesia, exacerbation of psychiatric symptoms, and gastrointestinal symptoms. Furthermore, entacapone requires frequent administration and dose adjustment, imposing a burden on patients, which has been a problem.

Selegiline has been shown to improve OFF-period symptoms, but its effect in reducing OFF time has not been demonstrated clearly. Regarding safety, as selegiline increases brain dopamine concentrations by acting centrally, it is likely to cause or exacerbate L-DOPA-induced peak-dose dyskinesias and the use of selegiline should be avoided if a patient has dyskinesia. Selegiline is contraindicated for use with antidepressants, which are expected to be used in patients with Parkinson's disease.

Zonisamide has been shown to improve motor function (improvement in the UPDRS part III score) at the approved dose of 25 mg, but its effect in reducing OFF time has not been demonstrated.

In light of the above-mentioned characteristics and problems of other anti-PD medications that are used for the treatment of wearing-off, istradefylline, which has been shown to reduce OFF time, which is considered important in the treatment of wearing-off, should be used in preference to selegiline which improves OFF-period symptoms only and zonisamide which has not been shown to reduce OFF time. While entacapone, like istradefylline, has been shown to reduce OFF time, istradefylline can improve motor function in the ON state as well. Regarding safety, the main adverse reaction to istradefylline was dyskinesia, but most of the reported dyskinesias were mild or moderate in severity. Entacapone and selegiline potentiate dopaminergic side effects, whereas non-dopaminergic istradefylline can be used safely also in patients suffering from dopaminergic side effects. Moreover, as mentioned above, there were no major differences in the efficacy and safety of istradefylline with or without concomitant anti-PD medications in the clinical studies. Therefore,

istradefylline can be used in a broad range of patients on levodopa-containing preparations, including those with untreated motor complications and those receiving ≥ 1 other anti-PD medications.

PMDA considers as follows:

The applicant claimed that istradefylline is expected to be effective in improving motor function in the ON state as well as reducing OFF time. However, since both Japanese late phase II and phase III studies included patients on levodopa-containing preparations with wearing-off as a motor complication and the primary endpoint was the change in OFF time in both studies, the intended population and the efficacy primarily expected of istradefylline should be “a reduction in OFF time (an improvement of the wearing-off phenomenon)” in “patients with Parkinson’s disease on levodopa-containing preparations with wearing-off phenomena.”

Clinical studies of istradefylline evaluated the efficacy and safety of istradefylline also in patients receiving other anti-PD medications that are used for the treatment of the wearing-off phenomenon and it can be concluded that the efficacy and safety of istradefylline are not significantly affected by the type or number of concomitant medications used. Thus, istradefylline can be used in Parkinson’s disease patients on levodopa alone or levodopa and ≥ 1 other anti-PD medications with wearing-off phenomena. Istradefylline, an adenosine A_{2A} receptor antagonist, has a different mechanism of action from currently available medications and can become a new pharmacotherapeutic option for Parkinson’s disease.

However, as to the applicant’s claim that istradefylline should be used in preference to selegiline and zonisamide in the treatment of Parkinson’s disease patients with wearing-off phenomena, since an appropriate medication should be selected from among anti-PD medications including istradefylline based on their efficacy and safety profiles and the patient’s condition etc., istradefylline is not positioned as a drug to be used in preference to currently available medications. The clinical positioning of istradefylline will be finalized, taking also account of comments from the Expert Discussion.

4.(iii).B.(2) Efficacy

4.(iii).B.(2).1 Effect on the wearing-off phenomenon

(a) Inconsistent results across studies

Foreign placebo-controlled clinical studies (Study 6002-US-005*, Study 6002-US-006*, Study 6002-US-013, Study 6002-US-018, Study 6002-EU-007) failed to demonstrate the consistent efficacy of istradefylline in reducing OFF time (the primary endpoint). PMDA asked the applicant to explain its cause in view of differences in the study design and subject background etc. among the studies.

* Foreign phase II studies in levodopa/carbidopa-treated Parkinson’s disease patients with motor complications (Reference data)

The applicant responded as follows:

Among 5 foreign placebo-controlled confirmatory clinical studies, Study 6002-US-005, Study 6002-US-006, and Study 6002-US-013 met the primary efficacy endpoint while Study 6002-US-018 and Study 6002-EU-007 failed to confirm the efficacy of istradefylline. As Study 6002-EU-007 showed no significant difference between entacapone vs. placebo and the internal validity of the study was not established, interpreting the results of this study itself is difficult. This was because the study was conducted as a multinational study in 14 countries including Europe, South America, India, and Russia, and center differences arising from differences in the medical environment and language etc. among many countries (the *P*-value for center-by-treatment interaction for the change in the mean total hours of awake time per day spent in the OFF state was 0.037) could not be controlled. Study 6002-US-018 could not satisfactorily detect the efficacy of istradefylline because the change in the mean total hours of awake time per day spent in the OFF state in the placebo group was very large (−1.42 hours) in this study compared with the other 4 studies (−0.86 to −0.60 hours). The possible factors contributing to the large change in the placebo group of Study 6002-US-018 were investigated. As a result, it was inferred that a high probability of being assigned to istradefylline (75%) etc., which can lead to a high placebo effect and higher baseline values for the mean total hours of awake time per day spent in the OFF state in this study compared with the other 4 studies etc. may have affected the outcome, but a definite cause could not be identified.

PMDA asked the applicant to explain the reason for different results with respect to the effect of istradefylline in reducing OFF time between Japanese late phase II and phase III studies and 2 foreign confirmatory studies (Study 6002-US-018, Study 6002-EU-007) in view of differences in the study design and subject background and intrinsic and extrinsic ethnic differences, etc. between Japan and overseas.

The applicant responded as follows:

As described above, it was considered that Study 6002-EU-007 and Study 6002-US-018 failed to demonstrate the efficacy of istradefylline due to center differences or different placebo effects. Then, the study design and subject background were compared between Japanese late phase II and phase III studies and 2 foreign studies (Study 6002-US-018, Study 6002-EU-007) to discuss their impact on the reduction in OFF time. Although the key sections of the protocol (the inclusion and exclusion criteria, the endpoints, etc.) were almost common to the Japanese and foreign studies, the major difference in the protocol between the Japanese and foreign studies was the number of daily diaries assessed at each timepoint. The diaries during the 7-day period preceding each visit were assessed in the Japanese late phase II and phase III studies while the diaries on two consecutive days before each visit were assessed in the foreign studies. Diaries were used as the basis for calculating the primary endpoint of the change in the mean total hours of awake time per day spent in the OFF state for the Japanese studies and the primary endpoint of the change in the mean percentage of awake time per day spent in the OFF state for the foreign studies, and the number of daily diaries assessed directly affects the precision of the mean. In a Japanese early phase II study (Study 6002-0406), the diaries on two consecutive days were assessed as in the foreign studies, but the intra-subject variability in OFF time was large across all groups and it was considered difficult to assess with due precision the effect of istradefylline. Based on this finding, in the subsequent Japanese late phase II and phase III studies, the

number of daily diaries assessed was changed from 2 to 7 and the study design was improved to reduce the intra-individual variability. With the increased precision of the data obtained from patient diaries, the Japanese confirmatory studies successfully confirmed the efficacy of istradefylline consistently. Finally, the impact of intrinsic and extrinsic ethnic factors on the reduction in OFF time was discussed. As to intrinsic ethnic factors, the pharmacokinetics of istradefylline were similar between Japanese and foreign subjects and there were no major differences in the pharmacokinetics of istradefylline according to age or gender. As to extrinsic ethnic factors, although the mean daily levodopa doses in Study 6002-US-018 and Study 6002-EU-007 (623.7-639.9 mg/day) were higher than those in the Japanese late phase II and phase III studies (416.2-425.5 mg/day), since there is little pharmacokinetic interaction with levodopa/carbidopa and a subgroup analysis by levodopa dose (<450 mg/day or ≥ 450 mg/day) in Japanese placebo-controlled comparative studies showed that patients treated with istradefylline consistently demonstrated a greater reduction in the mean total hours of awake time per day spent in the OFF state than those treated with placebo, there should be no effect of the dose of levodopa. Also as to smoking that may affect the pharmacokinetics of istradefylline, as the proportion of smokers was very low and similar in the Japanese and foreign studies (Japanese late phase II study, 7.8%; Japanese phase III study, 5.5%; Study 6002-US-018, 4.5%; Study 6002-EU-007, 5.2%), the effect of smoking status should be small. As extrinsic ethnic factors, the epidemiology of Parkinson's disease, diagnostic criteria, or treatment guidelines are not substantially different between Japan and overseas. In conclusion, in terms of intrinsic and extrinsic ethnic factors, a factor causing different results with respect to the effect of istradefylline in reducing OFF time between Japan and overseas could not be identified.

PMDA considers as follows:

As the applicant discussed, despite the fact that there were no major differences in the intrinsic and extrinsic ethnic factors, patient background, or study design between Japan and overseas, foreign clinical studies failed to clearly demonstrate the efficacy of istradefylline with no consistent results with respect to the effect of istradefylline in reducing OFF time across different studies. However, in the Japanese clinical studies compared with the foreign clinical studies, the number of daily diaries assessed was increased with an aim of increasing the precision of data and furthermore, the Japanese studies were conducted under the system where the results were less likely to be affected by center differences, compared with Study 6002-EU-007 that failed to demonstrate efficacy. Thus, the effect of istradefylline in reducing OFF time may have been assessed more accurately in the Japanese late phase II and phase III studies compared with the foreign clinical studies. In addition, in the Japanese development program, two placebo-controlled, parallel-group, comparative studies of similar design were conducted and the reproducibility of efficacy can also be assessed. Therefore, the efficacy of istradefylline should be evaluated based primarily on the data from the Japanese late phase II and phase III studies.

(b) Clinical significance of clinical study results

The applicant provided a justification of primary endpoint selection (the change in the mean total hours of awake time per day spent in the OFF state) for Japanese late phase II and phase III studies as follows:

In the development of istradefylline in Japan, it was decided to evaluate the treatment effect of istradefylline on the wearing-off phenomenon in patients with advanced (levodopa-treated) Parkinson's disease because preceding foreign phase II studies (Study 6002-US-005, Study 6002-US-006) had suggested the treatment effect of istradefylline on the wearing-off phenomenon. OFF time was chosen as the primary endpoint for Japanese clinical studies since OFF time was considered the most direct and important measure of the effect on the wearing-off phenomenon and was used as an endpoint also in studies of other anti-PD medications. In foreign placebo-controlled clinical studies, the mean percentage of awake time per day spent in the OFF state was employed as the primary endpoint because of the possibility that the total hours of OFF time might be affected by the total hours of sleep time on that day (or the total hours of awake time). However, a Japanese early phase II study (Study 6002-0406) and foreign placebo-controlled clinical studies showed almost equal effect sizes of istradefylline (the difference between istradefylline and placebo) for the mean percentage of daily OFF time and the mean daily OFF time. In addition, the mean daily OFF time rather than the mean percentage of daily OFF time is easier to interpret into the size of the clinical effect. Therefore, the mean total hours of awake time per day spent in the OFF state was chosen as the primary endpoint for Japanese late phase II and phase III studies.

As istradefylline is intended for use in advanced Parkinson's disease patients with wearing-off phenomena and the change in OFF time in these patients is a validated endpoint in Japan and overseas, PMDA considers that primary endpoint selection is justified. PMDA asked the applicant to explain the clinical significance of the difference by treatment (istradefylline 40 mg minus placebo) for the change from baseline in the mean daily OFF time in a Japanese phase III study (least-squares mean, -0.74 hours), also in view of the reason that the treatment difference observed in the Japanese phase III study was smaller than expected at the time of planning the study (1 hour).

The applicant responded as follows:

At the time of planning a Japanese phase III study, there was a lack of consensus about the definition of clinically significant reduction in the mean daily OFF time. Thus, as the effect size of istradefylline, the treatment difference between istradefylline and placebo was estimated to be 1 hour based on the results of an US phase II study (Study 6002-US-005, the least-squares mean difference between istradefylline 40 mg and placebo, -1.1 hours; SD, 2.5 hours) and a Japanese late phase II study (the least-squares mean difference between istradefylline 40 mg and placebo, -0.9 hours; SD, 2.2 hours), which were conducted previously in Japan or overseas and had a similar design to the Japanese phase III study. The Japanese phase III study showed that the least-squares mean difference between istradefylline 40 mg and placebo for the change in the mean daily OFF time was -0.74 [95% CI, -1.27, -0.20] hours. Although it was difficult to identify the definite reason for the smaller than expected treatment difference, the effect sizes in other Japanese and foreign clinical studies assessing the reduction in OFF time were all smaller than the effect size in Study

6002-US-005. The expected effect of istradefylline may have been overestimated for the Japanese phase III study because the effect was estimated based on Study 6002-US-005.

Although the efficacy of istradefylline shown in the Japanese phase III study may be smaller than estimated at the time of planning the study, the applicant's view on the clinical significance of the treatment difference between istradefylline 40 mg and placebo for the change in the mean daily OFF time (-0.74 hours) is as follows:

As OFF time was recorded in diaries that track PD status at 30-minute increments in clinical studies, a reduction of 0.74 hours means that one or two more 30-minute periods per day were recorded as ON time in the istradefylline group compared with the placebo group. The results of secondary endpoint analyses suggested that almost all of the reduction in OFF time was replaced by an increase in ON time "without troublesome dyskinesia" (+0.81 hours), which is a beneficial time for patients, in the istradefylline group. Furthermore, concerning one of the secondary endpoints, Clinical Global Impression-improvement (CGI-I) (clinician-rated treatment response), the proportions of subjects rated as "much improved" or better were higher with istradefylline, i.e. 10.7% (13 of 122 subjects) in the placebo group, 20.8% (25 of 120 subjects) in the istradefylline 20 mg group, and 28.7% (35 of 122 subjects) in the 40 mg group. As described in the above 4.(iii).B.(1), the effect of istradefylline in reducing OFF time is not affected by the type or combination of concomitant anti-PD medications used. Therefore, also when istradefylline is used with other medications that improve OFF time or OFF-period symptoms (entacapone, selegiline, zonisamide), a further reduction in OFF time can be obtained. Based on the above, the effect of istradefylline in reducing OFF time confirmed in the Japanese phase III study is of clinical significance.

PMDA asked the applicant to explain the significance of offering istradefylline to clinical practice, in view of the usefulness of istradefylline compared with entacapone, a marketed drug that is used to reduce OFF time.

The applicant responded as follows:

In Study 6002-EU-007, istradefylline and entacapone were compared in a parallel-group design. However, the objective of this study was not a head-to-head comparison of the efficacy of istradefylline vs. entacapone, but a placebo-controlled evaluation of the efficacy of istradefylline and entacapone and the entacapone group was included to establish the internal validity of the study. However, there was no significant difference between entacapone and placebo for the change from baseline in the mean percentage of daily OFF time ($P = 0.064$), indicating that the internal validity of the study could not be established, and the results of the study should be interpreted cautiously. Thus, although the study showed a smaller reduction in the mean percentage of daily OFF time with istradefylline compared with entacapone, the superiority or inferiority of istradefylline to entacapone in terms of reducing OFF time can not be discussed based on this study. On the other hand, regarding safety, the incidence of dyskinesias, which are commonly reported with istradefylline and entacapone, seemed comparable for the two drugs. However, a further reduction in OFF time can be expected also when istradefylline is used with other dopaminergic anti-PD medications that reduce OFF time, e.g. entacapone; as shown in 4.(iii).B.(2).2), istradefylline has been shown to improve motor function as

measured by the UPDRS part III total score; and istradefylline is more convenient due to once-daily administration and no need to frequently administer it simultaneously with each dose of a levodopa preparation, unlike entacapone. Therefore, as a therapeutic drug for Parkinson's disease patients with wearing-off phenomena, istradefylline with a non-dopaminergic mechanism of action can offer new usefulness, which is not available in dopaminergic entacapone.

PMDA considers as follows:

The treatment differences between istradefylline 20 mg and placebo and between istradefylline 40 mg and placebo for the reduction in the mean daily OFF time observed in a Japanese phase III confirmatory study were both about 0.7 hours, being smaller than estimated at the time of planning the study. However, as both Japanese late phase II and phase III studies consistently demonstrated the superiority of istradefylline 20 and 40 mg over placebo for the change in the mean daily OFF time and the results of secondary endpoint analyses (the change in ON time and CGI-I, etc.) also supported the efficacy of istradefylline. Therefore, the effect of istradefylline in improving the wearing-off phenomenon shown in the Japanese clinical studies is of clinical significance. Although there are no data allowing for efficacy comparison between istradefylline and entacapone, the Japanese late phase II and phase III studies including patients receiving concomitant entacapone confirmed the effect of istradefylline in reducing OFF time, and the results of a subgroup analysis suggested that the efficacy of istradefylline is not affected by concomitant entacapone. Thus, istradefylline has clinical significance in terms of offering a new therapeutic option for the improvement of the wearing-off phenomenon. As to the applicant's claim that istradefylline is more convenient due to once-daily administration, it can not be said that istradefylline can offer new usefulness, which is not available in entacapone, because there are no data showing that once-daily istradefylline is more useful than istradefylline frequently administered simultaneously with each dose of a levodopa-containing preparation and how the pharmacological effects of istradefylline contribute to its treatment effects has not fully been elucidated.

4.(iii).B.(2).2) Improvement of motor function

The applicant explained the effect of istradefylline in improving motor function as follows:

The UPDRS part III total score was chosen as a secondary endpoint for Japanese late phase II and phase III studies and the treatment differences between istradefylline 20 mg and placebo and between istradefylline 40 mg and placebo for the change in the UPDRS part III total score were both -2.0 [-3.5, -0.4] (least-squares mean [95% CI]) in the Japanese late phase II study and the treatment differences between istradefylline 20 mg and placebo and between istradefylline 40 mg and placebo were -0.9 [-2.3, 0.4] and -2.0 [-3.4, -0.7], respectively, in the Japanese phase III study. Istradefylline 20 mg and 40 mg reduced the UPDRS part III total score and there was a significant difference between istradefylline 40 mg and placebo in both studies. In conclusion, istradefylline has been shown to improve motor function in the ON state as well as reduce OFF time as described in the above 1) and istradefylline is considered useful as an anti-PD medication that is effective in both the OFF and ON states.

PMDA considers as follows:

The UPDRS part III total score was a secondary endpoint and Japanese late phase II and phase III studies were not designed to evaluate the effect of istradefylline in improving motor function. However, as the superiority of istradefylline 40 mg over placebo in reducing the UPDRS part III total score has been demonstrated consistently, improvement of motor symptoms, which are the cardinal symptoms of Parkinson's disease, can also be expected in Parkinson's disease patients with wearing-off phenomena, i.e. the intended population for istradefylline. On the other hand, the effect of istradefylline 20 mg in improving motor function has not been demonstrated clearly as the Japanese phase III study failed to show the superiority of istradefylline 20 mg over placebo.

4.(iii).B.(3) Safety

4.(iii).B.(3).1 Dyskinesia

The applicant explained dyskinesias reported in Japanese clinical studies as follows:

In subjects treated with istradefylline in 4 Japanese studies in patients with Parkinson's disease, dyskinesia was the most commonly reported adverse event (18.2%, 118 of 649 subjects), but many of the reported dyskinesias were mild in severity and none were rated as severe. According to the pooled analysis from Japanese placebo-controlled comparative studies, the incidence of dyskinesias was 3.3% (9 of 275 subjects) in the placebo group, 11.4% (31 of 272 subjects) in the 20 mg group, and 10.1% (28 of 277 subjects) in the 40 mg group and although the incidence was higher in istradefylline-treated subjects compared with placebo-treated subjects, there were no differences between the 20 mg and 40 mg groups. Furthermore, the AUC₀₋₂₄ values of individual patients after administration of 20 or 40 mg of istradefylline were estimated using Bayesian methods and then the estimates at each dose level were stratified by the occurrence of dyskinesia and compared using scatter plots. As a result, there was no noteworthy concentration of data points for exposures at the time of dyskinesia at either dose level. Subgroup analyses were performed to assess the effects of extrinsic factors on safety. As a result, the incidence of dyskinesias was higher in the subgroup treated with concomitant entacapone than in the subgroup treated without concomitant entacapone, i.e. 28.0% (59 of 211 subjects) and 13.5% (59 of 438 subjects), respectively, but many of the dyskinesias reported in the subgroup treated with concomitant entacapone were mild in severity and there was no substantial difference in the incidence between subjects treated with and without concomitant entacapone. Therefore, it was considered that concomitant use of entacapone would not markedly increase safety concern.

With respect to analyses by concomitant entacapone use, PMDA asked the applicant to present the data by treatment group and then explain any differences in the risk of dyskinesia between patients treated with istradefylline with and without concomitant entacapone.

The applicant responded as follows:

According to the pooled analysis from Japanese placebo-controlled comparative studies, the incidences of dyskinesias were 14.5% (25 of 172 subjects) in subjects treated with istradefylline with concomitant entacapone, 0% (0 of 69 subjects) in subjects treated with placebo with concomitant entacapone, 9.0% (34 of 377 subjects) in subjects treated with istradefylline without concomitant entacapone, and 4.4% (9 of 206

subjects) in subjects treated with placebo without concomitant entacapone and the incidence of dyskinesias was higher in the istradefylline group than in the placebo group, regardless of concomitant use of entacapone. Therefore, it is considered that even without concomitant entacapone use, patients treated with istradefylline develop dyskinesia and concomitant use of istradefylline and entacapone further increases the incidence of dyskinesias. In order to discuss the effect of concomitant entacapone use on the incidence of dyskinesias in istradefylline-treated patients, the incidences of dyskinesias in patients treated with istradefylline with and without concomitant entacapone were further analyzed according to the presence or absence of dyskinesia at baseline* and the duration of Parkinson's disease (≤ 10 years, >10 years), which are considered associated with the development of dyskinesia. As a result, the following findings were obtained:

- (a) Regardless of the presence or absence of dyskinesia at baseline, the incidence of dyskinesias was higher in subjects treated with istradefylline with concomitant entacapone than in those without concomitant entacapone.
- (b) Regardless of concomitant use of entacapone, the incidence of dyskinesias was higher in subjects with dyskinesia at baseline than in those without dyskinesia at baseline.
- (c) Among subjects with ≤ 10 years of the duration of Parkinson's disease, the incidence of dyskinesias was higher in subjects treated with istradefylline with concomitant entacapone than in those without concomitant entacapone. Among subjects with >10 years of disease duration, there were no differences in the incidence of dyskinesias between subjects treated with istradefylline with and without concomitant entacapone.

The above findings indicate that the development of dyskinesia in patients treated with istradefylline with concomitant entacapone is associated with the control of the therapeutic effect at or near the threshold for the development of dyskinesia by the optimized treatment of wearing-off. On the other hand, regardless of concomitant use of entacapone, patients with dyskinesia before treatment with istradefylline (dyskinetic patients) are likely to develop dyskinesia following treatment with istradefylline and the incidence of dyskinesias may increase in these patients as in patients treated with concomitant entacapone. Based on the above, it will be advised in the important precautions section of the draft package insert that "since istradefylline may worsen existing dyskinesia in patients, istradefylline should be administered while carefully monitoring the patient's condition."

PMDA considers as follows:

As the incidence of dyskinesias was higher in the istradefylline group than in the placebo group in all Japanese placebo-controlled comparative studies, there is a risk of dyskinesia, requiring caution, also during treatment with non-dopaminergic istradefylline. However, since dyskinesias reported in 4 Japanese studies in patients with Parkinson's disease were mild or moderate in severity and all subjects with dyskinesia leading to istradefylline discontinuation recovered upon discontinuation of istradefylline, the risk of

* "Baseline" was defined as the screening period for subjects who initiated treatment with Istradefylline in placebo-controlled comparative studies (Study 6002-0406, Study 6002-0608, Study 6002-009) and as the screening period of Study 6002-009 for subjects who initiated treatment with Istradefylline in a long-term treatment study (Study 6002-010) and subjects were classified based on diaries at baseline.

dyskinesia is acceptable. The applicant's response (a precaution statement regarding the use of istradefylline in patients with dyskinesia will be included in the package insert) is appropriate. In addition, a close attention should be paid to the finding that concomitant use of istradefylline and entacapone increases the incidence of dyskinesias and a relevant precaution statement should be included in the package insert. The precaution statements regarding dyskinesia will be finalized, taking also account of comments from the Expert Discussion.

4.(iii).B.(3).2) Psychiatric disorders (hallucination, visual hallucination, etc.)

The applicant explained psychiatric disorders (hallucination, visual hallucination, etc.) reported in Japanese clinical studies as follows:

According to the pooled analysis from 4 Japanese studies in patients with Parkinson's disease, as serious adverse events, hallucination (0.5% [3 of 649 subjects]), anxiety (0.3% [2 of 649 subjects]), delirium (0.3% [2 of 649 subjects]), persecutory delusion (0.3% [2 of 649 subjects]), depression (0.2% [1 of 649 subjects]), visual hallucination (0.2% [1 of 649 subjects]), and anxiety disorder (0.2% [1 of 649 subjects]) occurred in istradefylline-treated subjects. It has been reported that the incidence of psychiatric disorders increases with the progression of Parkinson's disease and during long-term use of anti-PD medications and it is not easy to control the symptoms. Thus, psychiatric disorders reported in Japanese clinical studies will be listed as "clinically significant adverse reactions" in the precautions section of the package insert to alert physicians. According to the pooled analysis from Japanese placebo-controlled comparative studies, no major differences between istradefylline 20 mg and 40 mg were observed for the incidences of individual adverse events, whereas the incidence of adverse events in the MedDRA/J System Organ Class (SOC) "psychiatric disorders" was 5.5% (15 of 272 subjects) in the 20 mg group and 10.1% (28 of 277 subjects) in the 40 mg group and the incidence of those classified as "other significant adverse drug reactions"* also tended to be slightly higher in the 40 mg group, i.e. 1.1% (3 of 272 subjects) in the 20 mg group and 3.2% (9 of 277 subjects) in the 40 mg group. Of 25 subjects with no concurrent or prior history of hallucination or visual hallucination who newly developed hallucination or visual hallucination following treatment with istradefylline, 8 subjects had hallucination or visual hallucination that was considered by the investigator to be likely caused by factors other than istradefylline (the primary disease, other anti-PD medications, etc.). Thus, the details of the hallucinations or visual hallucinations reported by the remaining 17 subjects were reviewed. As a result, these were all similar to hallucinations or visual hallucinations generally observed in patients with Parkinson's disease and there were no symptoms unique to istradefylline.

PMDA considers as follows:

Psychiatric disorders were reported as serious adverse events in Japanese clinical studies though the incidences were not high. Thus, the applicant's response (the reported psychiatric disorders will be listed as "clinically significant adverse reactions" to alert physicians) is appropriate. As it has been suggested that istradefylline increases the incidence of psychiatric disorders dose-dependently, whether the risks of 40 mg

* Non-serious adverse drug reactions leading to "discontinuation," "dose reduction," "dose increase," or "interruption" of study drug or "dose reduction of other anti-PD medications."

of istradefylline are acceptable when compared with its benefits will be determined in “4.(iii).B.(5) Dosage and administration.” The precaution statement in the package insert will be finalized, taking also account of comments from the Expert Discussion.

4.(iii).B.(3).3) Adverse events listed in the warnings and precautions section of the package insert for dopaminergic agents

(a) Impulse control disorder

As impulse control disorder has been reported as an adverse drug reaction of istradefylline in Japanese clinical studies, PMDA asked the applicant to explain whether a caution about impulse control disorder needs to be included in the important precautions section of the package insert.

The applicant responded as follows:

In 4 Japanese studies in patients with Parkinson’s disease, 1 subject had impulse control disorder (gambling and excitement) as an adverse drug reaction of istradefylline. However, the investigator commented that “the event is presumably related to a dopamine agonist, but an involvement of istradefylline can not be denied.” In this case, impulse control disorder was strongly suspected of being related to concomitant use of a dopamine agonist. Since only 1 event of impulse control disorder was reported in clinical studies and furthermore, this event was strongly suspected of being related to a factor other than istradefylline, if the information collected during the post-marketing experience shows increased impulse control disorders associated with istradefylline, an additional pharmacovigilance plan will be considered. As this will be enough to manage the problem, a caution about impulse control disorder in the important precautions section of the package insert is unnecessary at present.

PMDA considers as follows:

Although 1 subject developed impulse control disorder for which a causal relationship to istradefylline could not be denied in Japanese clinical studies, since impulse control disorder is listed as “clinically significant adverse reactions” in the draft package insert to alert physicians, there is currently no evidence that istradefylline directly potentiates the effects of dopaminergic agents, and it is already common knowledge in the treatment of Parkinson’s disease that attention should be paid to the possible development of impulse control disorder in patients on levodopa preparations, the applicant’s explanation that a further caution in the package insert for istradefylline is unnecessary at present and an additional action will be considered based on post-marketing information is acceptable. A final conclusion will be made, taking also account of comments from the Expert Discussion.

(b) Sudden onset of sleep, orthostatic hypotension, somnolence, and dizziness/vertigo, etc.

As adverse drug reactions of istradefylline, sudden onset of sleep, orthostatic hypotension, somnolence, and dizziness/vertigo etc. have been reported in Japanese clinical studies. PMDA instructed the applicant to include the following precaution statement in the package insert: Patients should be cautioned against engaging in potentially hazardous activities.

The applicant responded as follows:

As adverse drug reactions that would potentially affect driving a car or operating machinery, sudden onset of sleep, sleep attacks, and loss of consciousness (1 subject each, 0.2%) and syncope (3 subjects, 0.5%) were reported, according to the pooled analysis from 4 Japanese studies in patients with Parkinson's disease. All individual events occurred in the very small number of subjects and were mild or moderate in severity and there is no definite evidence that istradefylline induced these events. However, besides the above adverse drug reactions, somnolence (18 subjects, 2.8%), orthostatic hypotension (8 subjects, 1.2%), and dizziness/vertigo (dizziness postural, dizziness, vertigo) (a total of 11 subjects, 1.7%) were reported and the possibility that these adverse drug reactions affect potentially hazardous activities, such as driving a car or operating machinery, can not be ruled out. Therefore, it will be stated in the important precautions section of the draft package insert that "since a sudden onset of sleep without warning, sleep attacks, orthostatic hypotension, somnolence, dizziness/vertigo, loss of consciousness, and syncope etc. may occur, patients should be cautioned against engaging in potentially hazardous activities requiring alertness, such as driving a car, operating machinery, or working in high places, during treatment with Istradefylline."

PMDA considers that the applicant's response is appropriate.

4.(iii).B.(3).4) Use in patients with hepatic impairment

The exposure of istradefylline in subjects with moderate hepatic impairment was estimated to be about 3-fold that in healthy adult subjects [see "4.(ii).B.(2) Pharmacokinetics in subjects with hepatic impairment"], only the safety of multiple-dose administration of up to 80 mg of istradefylline has been demonstrated in Japanese subjects, and istradefylline has been shown to increase the risk of psychiatric disorders dose-dependently. Thus, PMDA asked the applicant to present safety information from Japanese and foreign patients with hepatic impairment treated with istradefylline and then explain whether istradefylline needs to be contraindicated or istradefylline dose adjustment is required in patients with hepatic impairment, depending on the degree of impairment.

The applicant responded as follows:

In accordance with the exclusion criteria, subjects with clinically significant hepatic impairment did not participate in Japanese or foreign clinical studies. Based on the subject background information from Japanese placebo-controlled comparative studies, subjects with medical conditions related to hepatic impairment were identified and then the safety information from 25 subjects with medical conditions related to hepatic impairment who were receiving 20 or 40 mg of istradefylline (20 mg, 11 subjects; 40 mg, 14 subjects) was reviewed. As a result, there were no serious adverse drug reactions and no noteworthy differences according to the degree of hepatic impairment were observed. However, as the number of these subjects was limited and patients with clinically significant hepatic impairment were not included, no conclusion on the necessity of dose adjustment etc. could be reached. Therefore, based on the benefits and risks estimated from the exposure of istradefylline, whether istradefylline dose adjustment is required in patients with moderate hepatic impairment and whether istradefylline may be used in patients with severe hepatic impairment were determined.

(a) Moderate hepatic impairment

As mentioned above, the exposure of istradefylline in subjects with moderate hepatic impairment was estimated to be about 3-fold that in healthy adult subjects and the exposures at 20 to 40 mg in subjects with moderate hepatic impairment are estimated to be equivalent to 60 to 120 mg in healthy adult subjects. Japanese clinical studies demonstrated the efficacy of istradefylline in reducing OFF time at doses of 20 and 40 mg and also in improving the UPDRS part III score at a dose of 40 mg. On the other hand, foreign clinical studies evaluated the effect of 20, 40, and 60 mg of istradefylline in reducing OFF time, which showed that the effect was not dose-related. It is inferred from the exposure of istradefylline that 20 mg of istradefylline reduces OFF time and improves the UPDRS part III score (motor function) in patients with moderate hepatic impairment and no further benefit can be expected when the dose is increased to 40 mg. Thus, a dose increase to 40 mg is considered unnecessary in patients with moderate hepatic impairment from an efficacy point of view. On the other hand, regarding safety, as described in “4.(iii).B.(3).2) Psychiatric disorders (hallucination, visual hallucination, etc.),” it has been suggested that the risk of “psychiatric disorders” is higher in patients treated with 40 mg than in those treated with 20 mg. As the exposure at 20 mg in patients with moderate hepatic impairment is equivalent to 60 mg in patients with normal hepatic function, there is concern about an even higher risk of psychiatric disorders. However, in foreign placebo-controlled clinical studies, the incidences of psychiatric disorders were 16.8% (74 of 440 subjects) in the 40 mg group and 18.1% (28 of 155 subjects) in the 60 mg group and there were no major differences in the incidence between 40 mg and 60 mg. Based on the above, in patients with moderate hepatic impairment, 20 mg of istradefylline is expected to produce demonstrated effects and its safety risks are considered acceptable. Therefore, considering the risk-benefit balance, it will be stated in the precautions of dosage and administration section of the draft package insert that the maximum dosage of istradefylline is 20 mg once daily in patients with moderate hepatic impairment.

(b) Severe hepatic impairment

The exposure of istradefylline in patients with severe hepatic impairment may exceed the exposure in patients with moderate hepatic impairment and the exposure at 20 mg in patients with severe hepatic impairment may be equivalent to ≥ 60 mg in healthy adults. As mentioned above, the efficacy of istradefylline has been evaluated at doses up to 60 mg in Japan or overseas, but there are no data on the effect of istradefylline at exposure levels higher than that at 60 mg. Regarding safety, since dyskinesia, hallucination, and agitation were reported at a dose of 120 mg in foreign clinical studies and Japanese clinical studies have also suggested a slight increase in the risk of psychiatric disorders with increasing dose, an increase in the above-mentioned risk, a further increase in the incidence of other adverse drug reactions, and new adverse drug reactions previously undocumented may occur even at a dose of 20 mg in patients with severe hepatic impairment. For patients with severe hepatic impairment, currently available anti-PD medications (entacapone, zonisamide, selegiline) may be used and even if istradefylline can not be used, these therapeutic drugs may be chosen. Based on the above, it was concluded that the use of istradefylline in patients with severe hepatic impairment should be “contraindicated” in the draft package insert.

PMDA considers as follows:

Istradefylline is eliminated from the body primarily by liver metabolism and the exposure even at 20 mg in patients with moderate hepatic impairment is considered to exceed the exposure at 40 mg in patients with normal hepatic function. The benefits of >40 mg of istradefylline are not clear from an efficacy point of view and an increased risk with increasing dose has been suggested from the safety standpoint. Thus, the applicant's response (Considering the risk-benefit balance, the maximum dose of istradefylline is 20 mg in patients with moderate hepatic impairment) is appropriate. Since there is no clinical experience with istradefylline and safety is unknown in patients with severe hepatic impairment and the possibility of an even higher exposure in patients with severe hepatic impairment than in patients with moderate hepatic impairment can not be denied, the applicant's response (The use of istradefylline in patients with severe hepatic impairment is "contraindicated") is also appropriate. Precautions regarding the use of istradefylline in patients with hepatic impairment will be determined, taking also account of comments from the Expert Discussion.

4.(iii).B.(4) Indication

Under the proposed indication of "Parkinson's disease patients with motor complications," istradefylline might be used in inappropriate patients, i.e. patients with dyskinesia only but without apparent OFF-period symptoms in whom the efficacy of istradefylline has not been studied. Since the change in OFF time was chosen as the primary endpoint for a Japanese phase III study to confirm the efficacy of istradefylline, PMDA instructed the applicant to reconsider the indication.

Based on PMDA's instruction, the applicant responded that istradefylline will be indicated for "patients with Parkinson's disease on levodopa-containing preparations with 'wearing-off' phenomena."

Since Japanese late phase II and phase III studies included patients taking at least 3 doses and ≥ 300 mg of levodopa/DCI per day, PMDA asked the applicant to explain whether the use of istradefylline should be restricted depending on levodopa dose, etc. in the precautions of indications section of the package insert so that patients with apparent wearing-off phenomena despite optimized treatment with levodopa preparations (or dopamine agonists) for whom istradefylline is expected to be effective can be selected appropriately.

The applicant responded as follows:

Japanese late phase II and phase III studies included patients taking at least 3 doses and ≥ 300 mg of levodopa/DCI per day to ensure that patients with more apparent wearing-off phenomena were selected. However, as "Parkinson's Disease Treatment Guideline 2011" recommends also the use of dopamine agonists before levodopa-containing preparations, depending on the patient background, even patients taking <300 mg/day of a levodopa-containing preparation may have wearing-off phenomena in clinical practice. Therefore, levodopa doses will not be specified as a criterion for the use of istradefylline and it will be stated in the precautions of indications section of the package insert that istradefylline should be used in patients with wearing-off phenomena despite optimized treatment with levodopa-containing preparations or dopamine agonists.

PMDA considers as follows:

The applicant's response (Based on the patient populations in Japanese clinical studies and the expected efficacy of istradefylline, the indication will be changed to "patients with Parkinson's disease on levodopa-containing preparations with 'wearing-off' phenomena" and the intended population for istradefylline will be further defined in the precautions of indications section of the package insert) is appropriate. A final conclusion will be made, taking also account of comments from the Expert Discussion.

4.(iii).B.(5) Dosage and administration

The applicant explained the rationale for the proposed dosage and administration as follows:

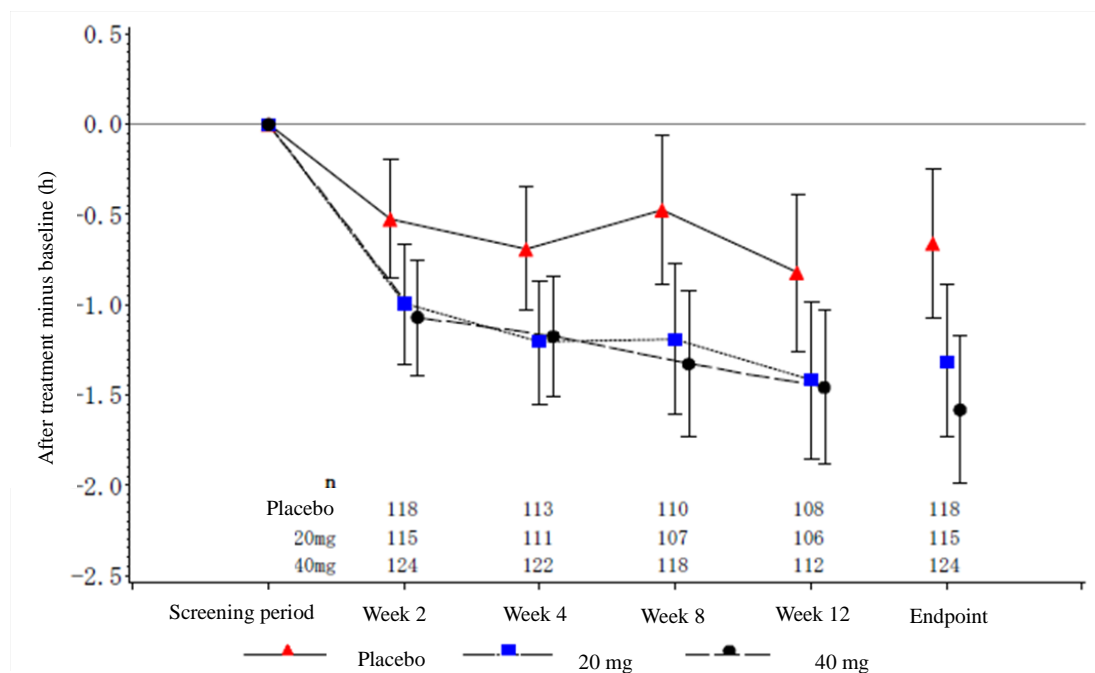
The $t_{1/2}$ after multiple-dose administration of 20 to 80 mg of istradefylline was 51.1 to 75.0 hours in a clinical pharmacology study, which was considered to allow once-daily administration. Thus, all 4 Japanese studies in patients with Parkinson's disease were conducted with once-daily administration. As a foreign late phase II study (Study 6005-US-005) had demonstrated the efficacy and safety of 40 mg prior to the conduct of a Japanese early phase II study (Study 6002-0406), 40 mg was chosen as the dose to be studied and 20 mg was also selected as a low dose group because the optimal dose had been estimated to be 20 to 40 mg in previous foreign clinical studies. The 20-mg and 40-mg doses were selected also for the subsequent Japanese clinical studies. According to the pooled analysis from Japanese placebo-controlled comparative studies, the least-squares mean differences between istradefylline 20 mg and placebo and between istradefylline 40 mg and placebo for the change from baseline to endpoint in the mean daily OFF time [95% CI] were -0.61 [-1.00, -0.23] hours and -0.79 [-1.16, -0.41] hours, respectively, showing a significant difference between either dose of istradefylline and placebo, but no major differences between the doses. However, in the dose titration period of a Japanese long-term treatment study (Study 6002-010), istradefylline was initiated at a dose of 20 mg and if the dose increase criteria were met at Week 4, the dose was allowed to be increased to 40 mg, and 55.2% of subjects (160 of 290 subjects) had their dose increased to 40 mg and the proportion of subjects who experienced a ≥ 1 -hour reduction in the mean daily OFF time from Week 4 to Week 8 was 30.0% (48 of 160 subjects) in subjects who had their dose increased to 40 mg, which was higher than 13.8% (18 of 130 subjects) in subjects who maintained on 20 mg. Therefore, the dose should be increased to 40 mg if the effect of istradefylline in reducing OFF time is inadequate. It is inferred from foreign clinical studies that the efficacy of istradefylline generally plateaus at doses >40 mg and furthermore, an apparent increase in the incidence of adverse events such as nausea has been observed at 60 mg compared with 20 to 40 mg.

PMDA considers as follows:

The effects of 20 mg and 40 mg of istradefylline on the primary endpoint of the change in OFF time were similar at endpoint and earlier timepoints in Japanese late phase II and phase III studies [see Figure 2 and Figure 3]. Due to the open-label design and differences in the patient background, it is difficult to say that subjects who maintained on 20 mg and subjects who had their dose increased to 40 mg in a Japanese long-term treatment study can be compared appropriately. Thus, it can not be said that a greater reduction in OFF time at 40 mg than at 20 mg has been suggested. Given that it has been suggested that the risk of psychiatric disorders is higher in patients treated with 40 mg than in those treated with 20 mg [see "4.(iii).B.(3).2)

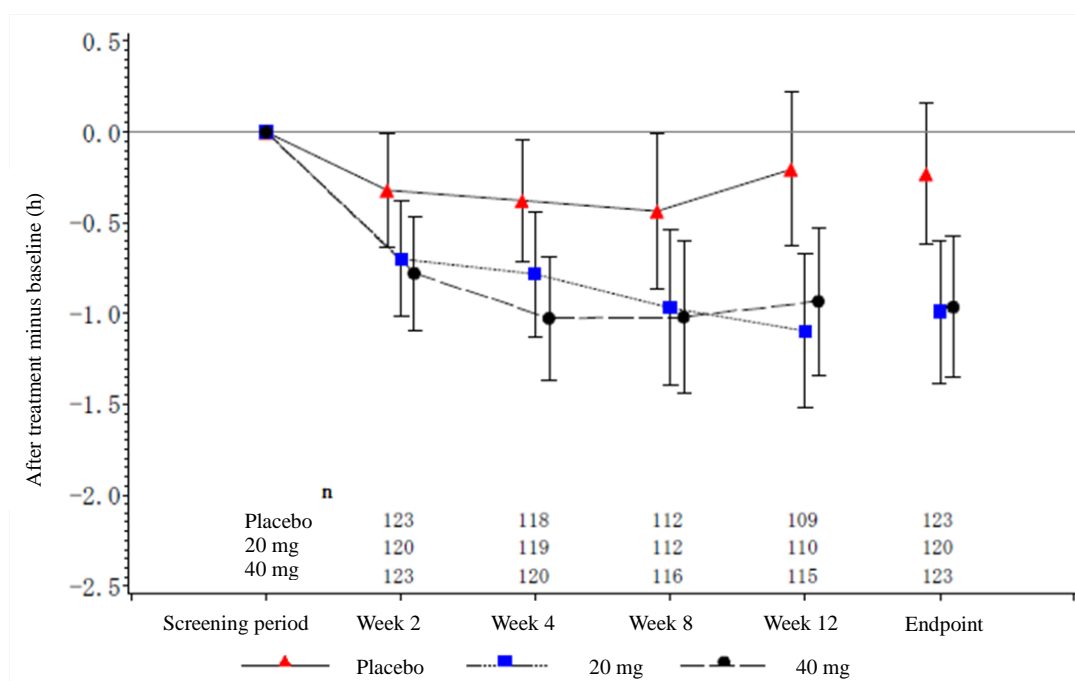
Psychiatric disorders (hallucination, visual hallucination, etc.)”], if a 40-mg dose is also recommended, who can benefit from 40 mg of istradefylline should be defined.

Based on the above, PMDA asked the applicant to provide a justification for dosage and administration of istradefylline again and explain appropriate precautions.



Baseline: Screening period, n: no. of subjects

Figure 2. Change in OFF time over time in Japanese late phase II study (Least-squares mean [95% CI])
(Adapted from Attached document 5.3.5.1-3 Figure 11.4.1.1-1)



Baseline: Screening period, n: no. of subjects

Figure 3. Change in OFF time over time in Japanese phase III study (Least-squares mean [95% CI])
(Adapted from Attached document 5.3.5.1-5 Figure 11.4.1.1.1-1)

The applicant responded as follows:

As the effects of 20 mg and 40 mg of istradefylline in reducing OFF time were similar in Japanese late phase II and phase III studies, the clinical usefulness of an increased dose of 40 mg in terms of reducing OFF time is not clear. On the other hand, regarding safety, as described above, according to the pooled analysis from Japanese placebo-controlled comparative studies, there were no major differences in the incidences of individual adverse events between 20 mg and 40 mg, whereas the incidence of psychiatric disorders tended to be slightly higher in the 40 mg group than in the 20 mg group. Therefore, the recommended dose of istradefylline should be 20 mg (the low dose) once daily.

However, it should also be taken into consideration that not only a reduction in OFF time, but also improvement in motor function in the ON state are needed by some Parkinson's disease patients with wearing-off phenomena, i.e. the intended population for istradefylline. As the effect of 40 mg of istradefylline in reducing the UPDRS part III total score in the ON state (improvement in motor function) was reproduced in the Japanese late phase II and phase III studies, not only a reduction in OFF time but also improvement in motor function in the ON state can be expected in Parkinson's disease patients with wearing-off phenomena, which has significant meaning for the treatment of Parkinson's disease patients with wearing-off phenomena.

Patients with wearing-off phenomena whose activities of daily living in the ON state are affected to a certain extent and who require treatment were defined as those with postural instability, i.e. modified Hoehn & Yahr stages ≥ 2.5 (in the ON state) and subjects in Japanese placebo-controlled comparative studies were divided according to modified Hoehn & Yahr stage (in the ON state) at baseline: stages ≥ 2.5 (2.5-4) (148 subjects in the placebo group, 154 subjects in the 20 mg group, 149 subjects in the 40 mg group) and stages ≤ 2 (0-2) (123 subjects, 112 subjects, and 126 subjects, respectively) and a subgroup analysis of motor function (UPDRS part III) in the ON state was performed. As a result, in the subgroup with stages ≤ 2 at baseline, the least-squares mean changes in the UPDRS part III total score [95% CI] in the placebo, 20 mg, and 40 mg groups were -3.1 [$-4.2, -2.0$], -4.7 [$-5.9, -3.5$], and -4.5 [$-5.6, -3.4$], respectively, and there were no differences in the reduction in the UPDRS part III total score between 20 mg and 40 mg. On the other hand, in the subgroup with stages ≥ 2.5 at baseline, the least-squares mean changes were -2.9 [$-3.9, -1.9$], -4.2 [$-5.2, -3.2$], and -5.5 [$-6.5, -4.5$], respectively, and the reduction in the UPDRS part III total score was greater at 40 mg than at 20 mg. A subgroup analysis of the incidence of adverse drug reactions was also performed in the same manner. As a result, there were no major differences in the incidence of adverse drug reactions between the doses in the subgroup with stages ≥ 2.5 at baseline and no major differences between the doses were observed also for the incidence of "psychiatric disorders," i.e. 1.3% (2 of 150 subjects) in the placebo group, 5.1% (8 of 157 subjects) in the 20 mg group, and 6.0% (9 of 150 subjects) in the 40 mg group. Therefore, it was considered that an increased dose of 40 mg is unlikely to increase risk markedly in patients with modified Hoehn & Yahr stages ≥ 2.5 (in the ON state).

Based on the above, since patients whose activities of daily living in the ON state were affected and who required treatment (modified Hoehn & Yahr stages ≥ 2.5 [in the ON state]) benefited from an increased dose of 40 mg of istradefylline and an increased dose of 40 mg is unlikely to increase risk markedly in these patients, it is considered that the benefits outweigh the risks and an increased dose of 40 mg is meaningful. In light of these points, it will be stated in the precautions of dosage and administration section of the draft package insert that “An increased dose of istradefylline is expected to improve motor function in the ON state. The dose should be increased in patients who need improvement in motor function in the ON state in addition to treatment of wearing-off (modified Hoehn & Yahr stages ≥ 2.5 , etc.).”

PMDA considers as follows:

The primary efficacy of istradefylline demonstrated in clinical studies is a reduction in OFF time in Parkinson’s disease patients with wearing-off phenomena. It can be concluded that placebo-controlled parallel-group comparative studies have demonstrated the effect of 20 mg in reducing OFF time. While there were no differences in the reduction in OFF time between 20 mg and 40 mg, the risk of psychiatric disorders may be higher at 40 mg. Taking account of these findings, the selection of 20 mg once daily as the recommended dose of istradefylline is justified.

As described above, a greater reduction in OFF time at 40 mg than at 20 mg has not been demonstrated and the risk may be higher at 40 mg than at 20 mg. Thus, the clinical significance of a dose increase is unclear when focusing on the reduction in OFF time only. However, the applicant’s claim that the improvement in motor function is also important for patients with Parkinson’s disease is understandable. Subjects treated with 40 mg consistently demonstrated a significant reduction in the UPDRS part III total score compared with placebo-treated subjects though the UPDRS part III total score was the secondary endpoint, and a significant difference between 40 mg and placebo has been demonstrated also for the reduction in OFF time. Therefore, offering a 40-mg dose in clinical practice is of significance though caution is required due to the risk of psychiatric disorders. Based on the results of post-hoc subgroup analyses, the applicant suggested that patients who need improvement in motor function in the ON state should be defined as those with modified Hoehn & Yahr stages ≥ 2.5 . Although it is appropriate to advise that a dose increase should be considered after understanding that the expected benefit of the 40-mg dose is the improvement in motor function, it is difficult to say that the results of the subgroup analyses presented by the applicant serve as clear evidence supporting the suggestion that the 40-mg dose should be considered for “patients with modified Hoehn & Yahr stages ≥ 2.5 .”

Based on the above, the dosage and administration statement should be as shown below. A dose increase should be considered after understanding that the expected benefit of the 40-mg dose is the improvement in motor function. The statements in the dosage and administration and precautions of dosage and administration sections of the package insert will be finalized, taking also account of comments from the Expert Discussion.

[Dosage and administration]

The product should be used in combination with levodopa-containing preparations. The usual adult dosage is 20 mg of Istradefylline orally administered once daily. ~~The dose should be increased as appropriate according to symptoms,~~ and 40 mg of Istradefylline may be orally administered once daily.

(Underline denotes additions and strikethrough denotes deletion.)

4.(iii).B.(6) Post-marketing surveillance etc.

Considering that it is necessary to appropriately collect information regarding the risks identified in non-clinical and clinical studies and the safety and efficacy of istradefylline in patient groups in whom limited data are available from clinical studies via post-marketing surveillance, PMDA instructed the applicant to determine the information to be collected via post-marketing surveillance and the sample size required.

Concerning the draft post-marketing surveillance plan, the applicant responded as follows:

A post-marketing surveillance study with an observation period of 1 year and a sample size of 1000 will be conducted to confirm the long-term safety and efficacy of istradefylline in routine clinical settings. The occurrence of psychiatric symptoms will be investigated as a priority and the following information will also be collected: (a) safety and efficacy in patients treated with an increased dose of 40 mg, (b) the relationship between the dose used and safety/efficacy in smoking patients, (c) safety in patients with hepatic impairment, (d) safety in patients with ischemic heart disease (the occurrence of arrhythmia), (e) the safety (including the occurrence of dyskinesia) and efficacy of istradefylline in combination with other anti-PD medications (by concomitant medication), and (f) the safety and efficacy of istradefylline in combination with CYP3A4 substrate drugs or CYP3A4 inhibitors or P-glycoprotein substrate drugs. If the information on dependence or lung toxicity (alveolar proteinosis, diffuse alveolar damage, laboratory findings/clinical symptoms suggestive of alveolar proteinosis or diffuse alveolar damage) as an adverse event is obtained, a detailed investigation will be conducted as appropriate.

PMDA considers that the information to be collected via post-marketing surveillance presented by the applicant is appropriate, but the details will be finalized, taking also account of comments from the Expert Discussion.

III. Results of Compliance Assessment Concerning the Data Submitted in the New Drug Application and Conclusion by PMDA

1. PMDA's conclusion on the results of document-based GLP/GCP inspections and data integrity assessment

A document-based compliance inspection and data integrity assessment was conducted in accordance with the provisions of the Pharmaceutical Affairs Act for the data submitted in the new drug application. As a result, PMDA concluded that there should be no problem with conducting a regulatory review based on the submitted application documents.

2. PMDA's conclusion on the results of GCP on-site inspection

GCP on-site inspection took place in accordance with the provisions of the Pharmaceutical Affairs Act for the data submitted in the new drug application (5.3.5.1-3, 5.3.5.1-5, 5.3.5.2-1). As a result, failure to retain some of the records required to be maintained at the trial site and protocol deviations (non-compliance with the rules for study drug dose increase) were found at some trial sites. In addition, it was found that the sponsor had been late in submitting its periodic reports on serious adverse drug reactions etc. to the trial sites and had failed to check the records required to be maintained at the above trial sites. Although these findings requiring improvement were noted, PMDA concluded that the clinical studies as a whole were performed in compliance with GCP and there should be no problem with conducting a regulatory review based on the submitted application documents.

IV. Overall Evaluation

Based on the submitted data, the effect of istradefylline in reducing OFF time in patients with Parkinson's disease on levodopa-containing preparations with wearing-off phenomena has been demonstrated and its safety is considered acceptable in view of its observed benefits. Istradefylline with a novel mechanism of action involving adenosine A_{2A} receptor antagonism offers a new therapeutic option for Parkinson's disease patients with wearing-off phenomena and is considered of clinical significance. The indication, dosage and administration, and precaution statements in the package insert for istradefylline, etc. need to be further discussed. It is necessary to appropriately collect post-marketing information regarding the risks of psychiatric symptoms, dyskinesia, dependence, and lung toxicity etc., safety and efficacy in patients treated with 40 mg, and safety in smokers, patients with hepatic impairment, and patients with ischemic heart disease.

Istradefylline may be approved if it can be concluded based on comments from the Expert Discussion that there are no particular problems.

Review Report (2)

February 22, 2013

I. Product Submitted for Registration

[Brand name]	Nourias Tablets 20 mg
[Non-proprietary name]	Istradefylline
[Name of applicant]	Kyowa Hakko Kirin Co., Ltd.
[Date of application]	March 30, 2012

II. Content of the Review

The Expert Discussion and subsequent review by the Pharmaceuticals and Medical Devices Agency (PMDA) are outlined below. The expert advisors for the Expert Discussion were nominated based on their declarations etc. concerning the product submitted for registration, 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).

1. Clinical positioning of istradefylline

The following conclusions by PMDA were supported by the expert advisors:

The intended population and the efficacy primarily expected of istradefylline should be “a reduction in OFF time (an improvement of the wearing-off phenomenon)” in “patients with Parkinson’s disease on levodopa-containing preparations with wearing-off phenomena.” Istradefylline can be used in patients on levodopa alone or levodopa and ≥ 1 other anti-PD medications. Istradefylline, an adenosine A_{2A} receptor antagonist, has a different mechanism of action from currently available medications and can become a new pharmacotherapeutic option for Parkinson’s disease.

Although the applicant claimed that istradefylline should be used in preference to selegiline and zonisamide, the expert advisors agreed that there is no evidence supporting the use of istradefylline in preference to currently available medications. PMDA’s conclusion that an appropriate medication should be selected from among anti-PD medications including istradefylline based on their efficacy and safety profiles and the patient’s condition etc. was supported by the expert advisors.

2. Effect of istradefylline on the wearing-off phenomenon

The expert advisors commented that although no consistent results with respect to the efficacy of istradefylline across different foreign clinical studies were obtained, Japanese clinical studies were designed to assess OFF time in more details, which evaluated the efficacy of istradefylline appropriately. PMDA’s conclusion that the efficacy of istradefylline should be evaluated based primarily on the data from Japanese late phase II and phase III studies (Study 6002-0608, Study 6002-009) was supported by the expert advisors.

The following conclusion by PMDA was supported by the expert advisors:

The Japanese late phase II and phase III studies consistently demonstrated the efficacy of 20 mg and 40 mg of istradefylline on the primary endpoint of the change in the mean daily OFF time (the effect of istradefylline in reducing OFF time). In addition, the results of secondary endpoint analyses (the change in ON time and CGI-I, etc.) also supported the efficacy of istradefylline. Thus, the effect of istradefylline in improving the wearing-off phenomenon shown in the Japanese clinical studies is of clinical significance.

The expert advisors made the following comments as well:

Given that a statistically significant difference from placebo was achieved only after the studies were designed to assess OFF time in more details and the observed effect size was smaller than expected at the time of planning the study, the magnitude of the effect of istradefylline seems limited. Physicians should be aware that the point estimate for the treatment difference between istradefylline and placebo in the reduction in OFF time was approximately 0.7 hours and should not use istradefylline aimlessly if the effect of istradefylline can not be expected.

Based on the above discussion, PMDA concluded that it is necessary to appropriately provide information so that the use of istradefylline can be decided after understanding the expected efficacy of istradefylline [see “4. Indication” and “5. Dosage and administration”].

3. Safety

3.(1) Dyskinesia

The following conclusions by PMDA were supported by the expert advisors:

As the incidence of dyskinesias was higher in the istradefylline group than in the placebo group in all Japanese placebo-controlled comparative studies (Study 6002-0406, Study 6002-0608, Study 6002-009), there is a risk of dyskinesia, requiring caution, also during treatment with non-dopaminergic istradefylline. The applicant’s response (a precaution statement regarding the use of istradefylline in patients with dyskinesia will be included in the package insert) is appropriate. It should also be stated in the precautions section of the package insert that concomitant use of istradefylline and entacapone increases the incidence of dyskinesias.

Based on the above, PMDA instructed the applicant to list entacapone in the precautions for coadministration section of the package insert to caution that concomitant use of istradefylline and entacapone increases the risk of dyskinesia. The applicant responded appropriately.

3.(2) Psychiatric disorders (hallucination, visual hallucination, etc.)

The following conclusion by PMDA was supported by the expert advisors:

Since psychiatric disorders were reported as serious adverse events in Japanese clinical studies, the applicant’s response (the reported adverse drug reactions of psychiatric disorders will be listed as “clinically significant adverse reactions” in the package insert to alert physicians) is appropriate.

3.(3) Impulse control disorder

The following conclusion by PMDA was supported by the expert advisors:

Since impulse control disorder is listed as “clinically significant adverse reactions” in the draft package insert to alert physicians, there is currently no evidence that istradefylline directly potentiates the effects of dopaminergic agents, and it is already common knowledge in the treatment of Parkinson’s disease that attention should be paid to the possible development of impulse control disorder in patients on levodopa preparations, the applicant’s explanation that a further caution in the package insert for istradefylline is unnecessary at present and an additional action will be considered based on post-marketing information is acceptable.

3.(4) Use in patients with hepatic impairment

The following conclusion by PMDA was supported by the expert advisors:

The applicant’s response (Based on the estimated increase in the exposure of istradefylline in patients with moderate hepatic impairment, no benefits of >40 mg of istradefylline demonstrated from an efficacy point of view, and an increased risk with increased exposure, the maximum dose of istradefylline will be 20 mg in patients with moderate hepatic impairment) is appropriate.

Whether istradefylline may be used in patients with severe hepatic impairment was discussed at the Expert Discussion. Some expert advisors commented that unlike levodopa preparations, istradefylline is not essential and is used as an adjunct in the treatment of Parkinson’s disease and there is less need to dare to use istradefylline in patients with severe hepatic impairment for whom the risks associated with istradefylline can not be estimated. Meanwhile, others commented that although the concerns about an increase in the exposure of istradefylline and no clinical experience with istradefylline in patients with severe hepatic impairment are understood, as no clear risks have been observed in patients with severe hepatic impairment at present, there is no need to contraindicate istradefylline in these patients.

In response to the above comments, PMDA explained as follows:

There is no clinical experience with istradefylline in patients with severe hepatic impairment and furthermore, it is difficult to predict the extent of increase in exposure and associated increased risk in these patients treated with istradefylline. Taking account of the comment from the expert advisors that istradefylline is not essential in the treatment of Parkinson’s disease, there are no benefits of using istradefylline in patients with severe hepatic impairment. Patients who are inappropriate for istradefylline should be listed in the contraindications section of the package insert and istradefylline should be contraindicated in patients with severe hepatic impairment.

After discussions, finally, it was decided to contraindicate istradefylline in patients with severe hepatic impairment and the understanding of the expert advisors was gained.

Since the expert advisors also commented that contraindicating istradefylline in patients with severe hepatic impairment just because of no clinical experience does not convey the above intention, PMDA instructed the

applicant to include the following information in the contraindications section of the package insert: Istradefylline is metabolized by the liver and the blood concentrations may be increased in patients with hepatic impairment. The applicant responded appropriately.

3.(5) CYP3A4- or P-glycoprotein-mediated drug interaction

The following conclusion by PMDA was supported by the expert advisors:

The applicant's responses (drugs that are CYP3A4 or P-glycoprotein substrates will be listed in the precautions for coadministration section of the package insert and the extent of increase in the plasma concentration of each drug observed in istradefylline drug interaction studies will be presented in the package insert) are appropriate.

The expert advisors commented as follows:

As atorvastatin is also a substrate of organic anion transporter OATP1B1, the increase in the blood concentration of atorvastatin when coadministered with istradefylline may have been due to OATP1B1 inhibition by istradefylline.

In response to the above comment, PMDA explained as follows:

The effect of istradefylline on OATP1B1 has not been investigated in *in vitro* studies. Since a drug interaction study with atorvastatin showed no clear decreases in the blood concentrations of atorvastatin metabolites formed by CYP3A4, the possibility that the increase in the blood concentration of atorvastatin was related to OATP1B1 inhibition by istradefylline can not be ruled out. However, istradefylline increased the exposure of atorvastatin about 1.5-fold, which was not great compared with the increase in the exposure of atorvastatin (an 8.7-fold increase) when coadministered with an OATP1B1 inhibitor, cyclosporine, which is listed in the precautions for coadministration section of the package insert for atorvastatin. Therefore, even if istradefylline inhibits OATP1B1, the degree of inhibition should not be great and istradefylline is unlikely to cause clinically relevant OATP1B1 inhibition. Regarding the safety of istradefylline in combination with other drugs including OATP1B1 substrate drugs, post-marketing information will be collected and if potential interactions are reported, an additional action will be considered as appropriate.

The above explanation by PMDA was supported by the expert advisors.

The following conclusion by PMDA was also supported by the expert advisors:

Based on the results of an istradefylline drug interaction study with ketoconazole, the dose of istradefylline should not exceed 20 mg when istradefylline is used concomitantly with potent CYP3A4 inhibitors like ketoconazole.

Based on the above discussions, PMDA instructed the applicant to include appropriate precaution statements in the precautions of dosage and administration section and the drug interactions section of the package insert. The applicant responded appropriately.

3.(6) Risks identified in non-clinical studies

1) Risk of arrhythmia during myocardial ischemia

PMDA concluded as follows:

It can not be said that a safety pharmacology study denied the possibility that istradefylline exacerbates arrhythmias during acute myocardial infarction and patients with clinically significant illness of any organ system, e.g. the heart (including serious abnormal findings in the ECG at screening) were excluded from Japanese clinical studies. Thus, it is necessary to caution about the risk of exacerbation of arrhythmias by istradefylline during myocardial ischemia in the package insert.

The expert advisors commented as follows and supported the conclusion by PMDA:

Once ventricular tachycardia occurs, it may lead to sudden death. Thus, the package insert should contain adequate caution statement.

Based on the above, PMDA instructed the applicant to list patients with ischemic heart disease in the careful administration section of the package insert and the applicant responded appropriately.

2) Lung effects

PMDA's conclusion that the package insert should contain adequate caution statement regarding the lung effects observed in toxicity studies was supported by the expert advisors and the expert advisors agreed that the applicant's response that the observed lung effects will be mentioned in the important precautions section and the other precautions section of the package insert is appropriate.

3) Dependence

The following conclusion by PMDA was supported by the expert advisors:

As the possibility that istradefylline causes psychological dependence in humans can not be ruled out, the package insert should caution that psychological dependence (a reinforcing effect) was observed in a non-clinical study, and risk management in clinical practice is required.

The expert advisors commented that the information on the occurrence of findings suggestive of dependence and the patient's condition after withdrawal of istradefylline, collected via post-marketing surveillance, would be useful.

4. Indication

The following conclusion by PMDA was discussed at the Expert Discussion:

The applicant's response (Based on the patient populations in Japanese clinical studies and the expected efficacy of istradefylline, the proposed indication will be changed to "patients with Parkinson's disease on levodopa-containing preparations with 'wearing-off' phenomena") is appropriate.

The expert advisors commented as follows:

Under the above indication proposed by the applicant, it may not be clearly understood that istradefylline should be used in expectation of an improvement of the wearing-off phenomenon and physicians might use istradefylline aimlessly without clearly being aware of the efficacy primarily expected of istradefylline. In order to avoid aimless administration to patients for whom istradefylline is not expected to be effective, it should be made clear in the indication statement that istradefylline is a drug intended to improve the wearing-off phenomenon (reduce OFF time). Concerning the statement in the precautions of indications section of the package insert, as the wording of “optimized treatment” with levodopa-containing preparations or dopamine agonists, proposed by the applicant, is abstract and may be interpreted in various ways, the word “optimized treatment” should be described more specifically.

Based on the above discussions, PMDA concluded that the indication statement and the statement in the precautions of indications section should be as shown below.

[Indication]

Improvement of the “wearing-off” phenomenon in patients with Parkinson’s disease on levodopa-containing preparations ~~with motor complications~~

[Precautions of indications]

The product should be used in patients with “wearing-off” phenomena despite adjustment of dose and dosing frequency of levodopa-containing preparations.

(Underline denotes additions and strikethrough denotes deletion)

5. Dosage and administration

The following conclusions by PMDA were supported by the expert advisors:

It can be concluded that Japanese late phase II and phase III studies have demonstrated the effect of istradefylline 20 mg in reducing OFF time and while there were no differences in the reduction in OFF time between 20 mg and 40 mg, the risk of psychiatric disorders may be higher at 40 mg. Taking account of these findings, the selection of 20 mg once daily as the recommended dose of istradefylline is justified.

In the Japanese late phase II and phase III studies, subjects treated with 40 mg consistently demonstrated a significant reduction in the Unified Parkinson’s disease rating scale (UPDRS) part III total score compared with placebo-treated subjects though the UPDRS part III total score was the secondary endpoint, and a significant difference between 40 mg and placebo was demonstrated also for the reduction in OFF time. Therefore, offering a 40-mg dose in clinical practice is of significance as long as the package insert advises that a dose increase should be considered after understanding that the expected benefit of the 40-mg dose compared with the 20-mg dose is the improvement in motor function.

The expert advisors commented as follows:

The package insert should clearly state that the expected benefit of 40 mg is the improvement in motor function. The package insert should also caution that no further reduction in OFF time was observed at 40 mg as compared with 20 mg and that the incidence of adverse events of psychiatric disorders may increase with increasing dose, so that whether patients should be treated with 40 mg of istradefylline can be determined appropriately.

Based on the above, PMDA concluded that the dosage and administration statement and the statements in the precautions of dosage and administration section should be as shown below. PMDA instructed the applicant to add the information on the incidence of adverse events of psychiatric disorders by dose level in Japanese clinical studies to the package insert and the applicant responded appropriately.

[Dosage and administration]

The product should be used in combination with levodopa-containing preparations. The usual adult dosage is 20 mg of Istradefylline orally administered once daily. ~~The dose should be increased as appropriate according to symptoms, and~~ 40 mg of Istradefylline may be orally administered once daily.

[Precautions of dosage and administration]

1. Istradefylline 40 mg may be orally administered once daily in expectation of an improvement of motor function in the “ON” state. Note that patients treated with 40 mg of Istradefylline did not demonstrate a greater reduction in “OFF” time than those treated with 20 mg (see “Clinical Studies”).

2. The dose of Istradefylline should not exceed 20 mg once daily in the following patients because blood concentrations of Istradefylline may be increased.

- Patients with moderate hepatic impairment (see “Careful Administration” and “Pharmacokinetics”)
- Patients receiving potent CYP3A4 inhibitors (see “Drug Interactions” and “Pharmacokinetics”)

(Underline denotes additions and strikethrough denotes deletion.)

6. Post-marketing surveillance etc.

The following conclusion by PMDA was supported by the expert advisors:

The information to be collected via post-marketing surveillance presented by the applicant is appropriate.

The expert advisors commented as follows:

The information on safety in patients with respiratory disorders and the occurrence of blood creatine phosphokinase (CK) increased, which occurred at a high incidence in clinical studies, and adverse events associated with blood CK increased (rhabdomyolysis, neuroleptic malignant syndrome, etc.) and the patient's condition at onset of these events (severity of Parkinson's disease, the dose of istradefylline, concomitant medications) should also be collected. The items for assessment of efficacy for treatment of motor dysfunction and the wearing-off phenomenon should also be included.

Based on the above, PMDA instructed the applicant to modify the post-marketing surveillance plan so that the above information can be collected appropriately and the applicant responded that the information will be collected in line with PMDA's instruction.

PMDA concluded as follows:

Although the details of a post-marketing surveillance protocol etc. will need to be determined, the draft post-marketing surveillance plan is largely appropriate.

III. Overall Evaluation

As a result of the above review, PMDA concludes that istradefylline may be approved for the following indication and dosage and administration. The re-examination period should be 8 years, neither the drug substance nor the drug product is classified as a poisonous drug or a powerful drug, and the product is not classified as a biological product or a specified biological product.

[Indication]

Improvement of the “wearing-off” phenomenon in patients with Parkinson's disease on levodopa-containing preparations

[Dosage and administration]

The product should be used in combination with levodopa-containing preparations. The usual adult dosage is 20 mg of Istradefylline orally administered once daily. According to symptoms, 40 mg of Istradefylline may be orally administered once daily.