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]	Metreleptin for Subcutaneous Injection 11.25 mg "Shionogi"
[Non-proprietary name]	Metreleptin (Genetical Recombination) (JAN*)
[Applicant]	Shionogi & Co., Ltd.
[Date of application]	July 27, 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 presented 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 10 years, and neither the drug substance nor the drug product is classified as a poisonous drug or a powerful drug.

[Conditions for approval]
The applicant is required to conduct a drug use-results survey involving all patients treated with the product after the market launch until data from a certain number of patients have been accumulated in order to grasp the demographic information of the treated patients, since the product has been studied in only a limited number of patients in the Japanese clinical studies. At the same time, data on the safety and efficacy of the product should be collected without delay and necessary measures should be taken to ensure proper use of the product.

**Japanese Accepted Name (modified INN)*

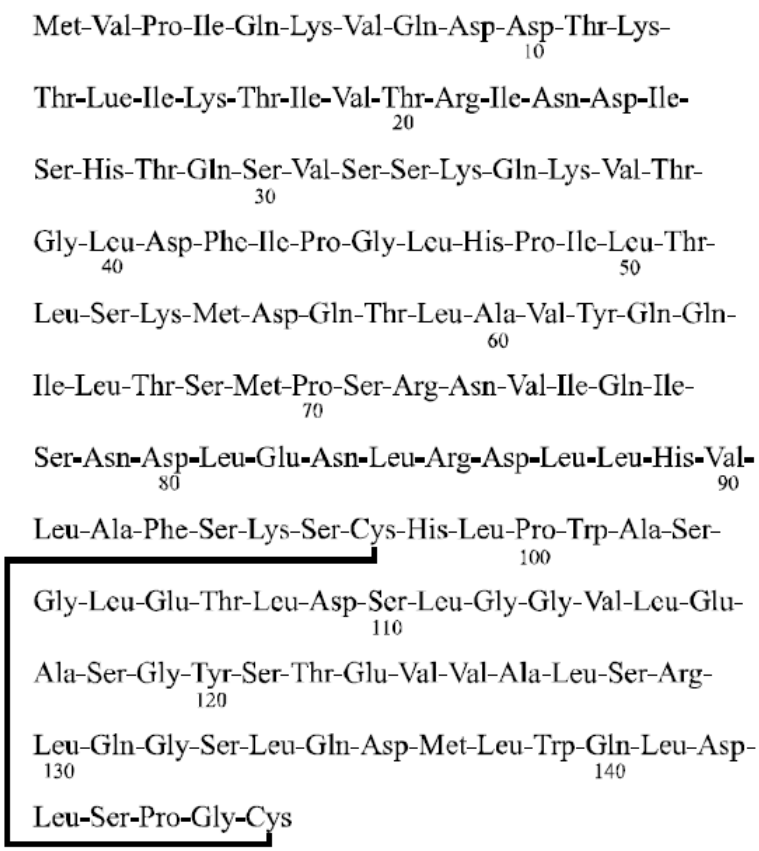
Review Report

February 19, 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]	Metreleptin for Subcutaneous Injection 11.25 mg "Shionogi" (changed from Metreleptin for Subcutaneous Injection 11.3 mg "Shionogi")
[Non-proprietary name]	Metreleptin (Genetical Recombination)
[Name of applicant]	Shionogi & Co., Ltd.
[Date of application]	July 27, 2012
[Dosage form/Strength]	Lyophilized formulation for solution for injection: Each vial contains 11.25 mg of Metreleptin (Genetical Recombination).
[Application classification]	Prescription drug (1) Drug(s) with a new active ingredient

[Chemical structure]



Molecular formula: $C_{714}H_{1167}N_{191}O_{221}S_6$

Molecular weight: 16155.44

Chemical name:

Metreleptin is a recombinant N-methionyl human leptin consisting of 147 amino acid residues.

[Items warranting special mention] Orphan drug (Notification No.0613-1 from the Evaluation and Licensing Division, Pharmaceutical and Food Safety Bureau, MHLW, dated June 13, 2012)

[Reviewing office] Office of New Drug I

Review Results

February 19, 2013

[Brand name]	Metreleptin for Subcutaneous Injection 11.25 mg "Shionogi" (changed from Metreleptin for Subcutaneous Injection 11.3 mg "Shionogi")
[Non-proprietary name]	Metreleptin (Genetical Recombination)
[Name of applicant]	Shionogi & Co., Ltd.
[Date of application]	July 27, 2012
[Results of review]	Based on the submitted data, it is concluded that the efficacy of the product in the treatment of patients with lipodystrophy has been demonstrated and its safety is acceptable in view of its observed benefits. The safety and efficacy of the product need to be further investigated via post-marketing surveillance. As a result of its regulatory review, the Pharmaceuticals and Medical Devices Agency has concluded that the product may be approved for the indication and dosage and administration as shown below, with the following conditions.
[Indication]	Lipodystrophy
[Dosage and administration]	The usual dosage is 0.04 mg/kg as metreleptin for male patients, 0.06 mg/kg for female patients aged <18 years, and 0.08 mg/kg for female patients aged ≥18 years, administered subcutaneously once daily. Treatment should be initiated at a dose of 0.02 mg/kg, 0.03 mg/kg, and 0.04 mg/kg, respectively, and the dose should be increased up to the dose level specified above within about a month. The dose may be adjusted according to the patient's symptoms.
[Conditions for approval]	The applicant is required to conduct a drug use-results survey involving all patients treated with the product after the market launch until data from a certain number of patients have been accumulated in order to grasp the demographic information of the treated patients, since the product has been studied in only a limited number of patients in the Japanese clinical studies. At the same time, data on the safety and efficacy of the product should be collected without delay and necessary measures should be taken to ensure proper use of the product.

Review Report (1)

January 11, 2013

I. Product Submitted for Registration

[Brand name] Metreleptin for Subcutaneous Injection 11.3 mg "Shionogi"

[Non-proprietary name] Metreleptin (Genetical Recombination)

[Name of applicant] Shionogi & Co., Ltd.

[Date of application] July 27, 2012

[Dosage form/Strength] Lyophilized formulation for solution for injection: Each vial contains 11.25 mg of Metreleptin (Genetical Recombination).

[Proposed indication] Improvement of hyperglycemia and hypertriglyceridemia in the following disease:

Lipodystrophy

[Proposed dosage and administration]

The usual dosage is 0.04 mg/kg as metreleptin for male patients, 0.06 mg/kg for female patients aged <18 years, and 0.08 mg/kg for female patients aged ≥18 years, administered subcutaneously once daily.

Treatment should be initiated at a dose of 0.02 mg/kg, 0.03 mg/kg, and 0.04 mg/kg, respectively, and the dose should be increased up to the dose level specified above within about a month.

The dose may be adjusted according to the patient's symptoms.

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

A summary of the submitted data and an outline of the 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.

The proposed product is a solution for subcutaneous injection containing Metreleptin (Genetical Recombination), an N-methionyl human leptin analog (hereinafter referred to as metreleptin).

Leptin, a hormone secreted primarily by adipocytes, is involved in the transmission of information on energy accumulation, to the central nervous system (Friedman JM, et al. *Nature*. 1998;395:763-70). By transmitting strong signals to suppress food intake through its direct effects on leptin receptors found mainly in the hypothalamus, leptin also improves energy expenditure, increases insulin sensitivity, and enhances lipid metabolism, and thereby plays an important role in glucose and lipid metabolism.[1]

[1] Halaas JL, et al. *Science*. 1995;269:543-6. Farooqi IS, et al. *Am J Clin Nutr*. 2009;89:980S-4S, Friedman JM. *Am J Clin Nutr.* 2009;89:973S-9S.

Lipodystrophy is characterized by loss of adipose tissue and classified into the following types: congenital generalized lipodystrophy, familial partial lipodystrophy, acquired generalized lipodystrophy, and acquired partial lipodystrophy (Chan JL, et al. *Endocr Pract.* 2010;16:310-23). The pathophysiology of lipodystrophy has not been clarified but excessive ectopic fat accumulation in the liver and skeletal muscles due to complete loss of adipose tissue or a marked decrease in adipose tissue is considered to be involved in increased insulin resistance (Simha V, et al. *Curr Opin Lipidol.* 2006:17;162-9). Congenital generalized lipodystrophy is considered to be caused by the lack of adipocytes due to mutations in the gene encoding 1-acyl-sn-glycerol-3-phospate acyltransferase (AGPAT2, an enzyme involved in the synthesis of triglycerides and phospholipids); and the genes encoding seipin and Caveolin 1, both of which are proteins involved in fat accumulation in adipose tissue. Familial partial lipodystrophy is characterized by partial loss of adipocytes due to mutations in the LMNA gene encoding a nuclear lamina protein (lamin A); the gene encoding peroxisome proliferator-activated receptor gamma (PPARγ), a transcription factor involved in adipocyte differentiation; and the gene encoding AKT2/protein kinase B (PKB), an enzyme for protein phosphorylation, which is involved in the insulin signal transduction pathway. Although the pathogenesis of acquired lipodystrophy is unknown, both generalized and partial forms of acquired lipodystrophy are considered to be associated with autoimmune diseases, resulting in loss of adipocytes. As shown in the above, a lack or partial loss of adipose tissue occurs in patients with lipodystrophy due to congenital and acquired factors, leading to a lack of adipocytokines including leptin. As a result, triglycerides accumulate in the liver, skeletal muscles and other organs, and thereby cause severe hepatic steatosis, hypertriglyceridemia, insulin resistance, etc., which results in elevated blood glucose levels.[2] Medical conditions related to metabolic abnormalities affecting long-term prognosis of patients with lipodystrophy include non-alcoholic steatohepatitis with subsequent cirrhosis, acute pancreatitis due to hypertriglyceridemia, and diabetic complications, hypertrophic cardiomyopathy, and atherosclerosis, which are caused by chronic hyperglycemia and hyperinsulinemia. In severe cases, non-alcoholic steatohepatitis can progress to cirrhosis, often leading to death. The natural course of lipodystrophy is not fully understood since its actual conditions have not been sufficiently studied in Japan and overseas, while it is reported that lipodystrophy is a refractory disease with an extremely poor prognosis and patients with lipodystrophy have an average life expectancy of 30 to 40 years. Almost no epidemiological data are available on the number of patients with lipodystrophy. However, according to the results from the surveys of 1559 endocrinologists of the Japan Endocrine Society in 2007, 31 patients are found to be affected by lipodystrophy, and the number of patients with lipodystrophy is estimated to be about 100 in Japan (Ebihara K, et al. *Journal of Japan Society for the Study of Obesity*. 2011;17:15-20). The proposed indication of the product is "Treatment of diabetes mellitus or dyslipidemia caused by lipodystrophy" and it has been designated as an orphan drug (Designation No. [24 yaku] 277).

A causal treatment of lipodystrophy is not available and no standard treatment has been established. As symptomatic therapy, patients are treated with dietary restrictions, antidiabetic drugs, or antihyperlipidemic drugs but these symptomatic therapies are often not effective for treating patients with severe lipodystrophy (Oral EA, et al. *Endocr Pract.* 2010;16:324-33). Although IGF-I preparation (mecasermin [genetical

[2] Chan JL, et al. *Endocr Pract.* 2010;16:310-23, Capeau J, et al. *Endocr Dev.* 2010;19:1-20

recombination]) has been approved for the indication of symptomatic therapy for lipoatrophic diabetes mellitus to regulate blood glucose levels, its therapeutic effects are limited (Ebihara K, et al. *Journal of Japan Society for the Study of Obesity*. 2011;17:15-20).

Clinical studies including the Japanese investigator-initiated trial (Study KUTR-003-1) have demonstrated the efficacy of the product in lipodystrophy treatment, and thus the applicant has filed a marketing application.

In foreign countries, metreleptin development was first started by Amgen Inc. (US) for the treatment of obesity and clinical studies were initiated in 19[redacted]. The United States National Institutes of Health (NIH) started clinical studies for clinical development of metreleptin for treating lipodystrophy in 2000 and it has been investigated under compassionate use. After in-licensing metreleptin from Amgen in 2006, Amylin Pharmaceuticals, LLC has conducted clinical studies of lipodystrophy treatment since 20[redacted]. Metreleptin was designated as an orphan drug for lipodystrophy in the US and the EU in 2001 and 2012, respectively.

A marketing application was submitted in [redacted] 20[redacted] in the US, but by July 2012, no countries have approved the drug product for the treatment of lipodystrophy.

2. Data relating to quality

2.A Summary of the submitted data

2.A.(1) Drug substance

2.A.(1).1) Preparation and control of cell substrates

A DNA fragment was chemically synthesized by the addition of the nucleotide sequence encoding methionine to the 5'-terminus of the nucleotide sequence encoding human leptin, and inserted into an expression vector to generate an expression construct. This expression construct was used to transform *Escherichia coli* (*E. coli*) cells. The seed stock, master cell bank (MCB), and working cell bank (WCB) were established from the transformed *E. coli* cells.

Tests were performed to characterize the MCB and WCB (DNA sequence analysis, plasmid retention, genotyping, test for absence of bacteria and fungi, and test for absence of bacteriophages), and all test results were within action limits. Appropriate storage conditions have been set for the MCB and WCB, and a new MCB or WCB will be generated as necessary.

2.A.(1).2) Manufacturing process

The manufacturing process of the drug substance comprises fermentation, inclusion body isolation, [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], [redacted] chromatography, [redacted] chromatography, [redacted] [redacted] chromatography, ultrafiltration/diafiltration, formulation, filtration, and filling. The fermentation process, inclusion body isolation, [redacted], [redacted] chromatography, [redacted] chromatography, formulation, filtration, and filling have been defined as critical process steps.

Validation of the commercial scale manufacturing process for the drug substance has demonstrated that each process step is adequately controlled.

2.A.(1).3) Adventitious agent safety evaluation

As biological materials, Bacto Tryptone and Trypticase Peptone produced from bovine milk using enzymes derived from the porcine pancreas were used in the fermentation process. All materials have been confirmed to meet the criteria for biological materials.

2.A.(1).4) Manufacturing process development (comparability)

Major changes in the manufacturing process for the development of the drug substance are as shown in Table 1.

Table 1. Major changes in the manufacturing process for the development of the drug substance

Development process	Changes
Phase I clinical batches to phase I scale-up clinical batches	・ Culture scale ・ Changes in conditions for [redacted], [redacted] chromatography, [redacted] chromatography, and ultrafiltration/diafiltration ・ Addition of [redacted] chromatography
Phase I scale-up clinical batches to phase II clinical batches	・ [redacted] method
Phase II clinical batches to phase III clinical batches	・ Culture scale ・ [redacted] method ・ Changes in conditions for [redacted] chromatography and [redacted] chromatography
Phase III clinical batches to 20[redacted] campaign batches	・ Use of [redacted] ・ Culture scale ・ Collection of bacterial cells and inclusion bodies ・ [redacted] scale ・ Separation of processes for [redacted] and [redacted] ・ Changes in conditions for [redacted] chromatography
20[redacted] campaign batches to validation batches	・ Culture scale

Following these changes in the manufacturing process, comparability studies on quality attributes were conducted and the comparability of the drug substances before and after the changes in the manufacturing process was confirmed.

2.A.(1).5) Characterization

(a) Structure/Composition

For the primary structure, amino acid sequence, disulfide bond, terminal amino acid sequence, and molecular weight were examined and the validity of theoretical sequence or theoretical values was supported by the results obtained.

The secondary structure and higher order structure were determined by far-ultraviolet circular dichroic spectroscopy, near-ultraviolet circular dichroic spectroscopy, fluorescence spectroscopy, size exclusion chromatography, and ultracentrifugal analysis.

(b) Physicochemical properties

The isoelectric point, extinction coefficient, and solubility were determined.

(c) Biological properties

The biological activity was determined using a cell proliferation bioassay.

(d) Product-related substances

[redacted] and [redacted] determined by reversed-phase liquid chromatography (RP-HPLC) were identified as product-related substances.

(e) Impurities

i) Process-related impurities

Host-derived proteins, DNA, endotoxins, and viable cells were identified as process-related impurities. These impurities have been demonstrated to be consistently removed through the manufacturing process. All of these impurities are controlled according to the drug substance specifications.

ii) Product-related impurities

[redacted], [redacted], [redacted], [redacted], [redacted], [redacted], [redacted], [redacted] oligomer and [redacted] non-dissociative oligomer were identified as product-related impurities. These impurities are controlled according to the drug substance specifications.

2.A.(1).6) Control of drug substance

The proposed specifications for the drug substance include content, description, identification (1) (SDS-polyacrylamide gel electrophoresis [SDS-PAGE]), identification (2) (peptide mapping), pH, oligomer (size exclusion chromatography [SEC-HPLC]), purity ([1] related substances I [SDS-PAGE], [2] related substances II [RP-HPLC], [3] non-dissociative oligomer [SEC-HPLC], [4] DNA [[redacted] [redacted]], [5] host-derived proteins [[redacted]]), bacterial endotoxins, total viable count, bioactivity (bioassay), and assay (ultraviolet-visible spectrophotometry).

2.A.(1).7) Stability of drug substance

A summary of the main stability studies of the drug substance is as shown in Table 2.

Table 2. Summary of the main stability studies of the drug substance

	Manufacturing process	Number of batches	Storage conditions	Type of storage	Testing frequency
Long-term testing	20[redacted] campaign batches[a)]	3	[redacted]°C	[redacted] bottle with a [redacted] cap	48 months
Accelerated testing	20[redacted] campaign batches[b)]	3	[redacted]°C		6 months

a) Long-term testing has been conducted for 3 drug substance validation batches for [redacted] months.
b) Accelerated testing has been conducted for 3 drug substance validation batches for [redacted] months.

No changes were observed in the long-term testing. In the accelerated testing, changes were observed for related substances II and content.

Photostability was not evaluated.

Based on the above, a shelf life of [redacted] months has been proposed for the drug substance when stored in a [redacted] bottle with a [redacted] cap at [redacted] °C, protected from light.

2.A.(2) Drug product

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

The drug product is a lyophilized formulation for solution for injection that contains 11.25 mg of drug substance in a vial. Each drug product contains the following excipients: glycine, purified sucrose, polysorbate 20, L-glutamic acid, and sodium hydroxide.

2.A.(2).2) Manufacturing process

The manufacturing process of the drug product comprises weighing and dissolution, sterile filtration and filling, freeze drying, sealing, and labeling and packaging. Sterile filtration and filling, freeze drying, and sealing have been defined as critical process steps.

Validation of the commercial scale manufacturing process for the drug product has demonstrated that each process step is adequately controlled.

2.A.(2).3) Manufacturing process development

In the course of development of the drug product, there was a formulation change (from a frozen formulation[3] to a lyophilized formulation), and the manufacturing site was also changed. The results from studies on quality attributes have indicated the comparability of pre- and post-change products.

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

The proposed specifications for the drug product include content, description, identification (1) (SDS-PAGE), identification (2) (peptide mapping), pH, oligomer (SEC-HPLC), purity ([1] appearance of solution, [2] related substances I [SDS-PAGE], [3] related substances II [RP-HPLC], and [4] non-dissociative oligomer [SEC-HPLC]), water content, uniformity of dosage unit, foreign insoluble matter, insoluble particulate matter, sterilization, bioactivity (bioassay), and assay (ultraviolet-visible spectrophotometry).

2.A.(2).5) Stability of drug product

A summary of the main stability studies of the drug product is as shown in Table 3.

[3] A frozen formulation is a solution for injection that is kept frozen for preservation. It was thawed before use.

Table 3. Summary of the main stability studies of the drug product

	Manufacturing process for the drug substance	Number of batches	Storage conditions	Type of storage	Testing frequency
Long-term testing	Phase III clinical batches	3	5 ± 3°C	Glass bottle	36-38 months
	20[redacted] campaign batches	2	5 ± 3°C		18/36 months[a)]
	Validation batches	1	5 ± 3°C		9 months[a)]
Accelerated testing	Phase III clinical batches	2	25 ± 2°C		6 months
	20[redacted] campaign batches	2	25 ± 2°C		6 months
	Validation batches	1	25 ± 2°C		6 months
Photostability testing	20[redacted] campaign batches	1	1,200,000 lux·hr (integrated illuminance) and 200 W·h/m^2 (total near-ultraviolet radiation energy)	Glass bottle and light-resistant glass bottle	-

a) Stability tests are on-going.

No apparent changes were observed in the long-term testing.

The accelerated testing showed a trend towards a slight increase in water content.

The photostability testing demonstrated an increase in related substances and a decrease in purity, indicating that the drug substance was photosensitive.

Based on the above, a shelf-life of 36 months has been proposed for the drug product when stored in a glass bottle at 2°C to 8°C, protected from light.

2.A.(3) Reference materials

The reference material is [redacted] and the proposed specifications include content, description, identification (1) (SDS-PAGE), identification (2) (peptide mapping), pH, oligomer (SEC-HPLC), purity ([1] related substances I [SDS-PAGE], [2] related substances II [RP-HPLC], and [3] non-dissociative oligomer [SEC-HPLC]), water content, bioactivity (bioassay), and assay (ultraviolet-visible spectrophotometry).

2.B Outline of the review by PMDA

Based on the submitted data and the following reviews, PMDA has concluded that the quality of the drug substance and the drug product is appropriately controlled.

Stability

PMDA asked the applicant to explain the justification for establishing the shelf life for the proposed drug product based on the results of the long-term testing using batches of the drug substance for the phase III clinical studies.

The applicant responded as follows:

Results from the analysis of quality attributes and stability studies have demonstrated the comparability of the drug substances between the phase III clinical batches and 20[redacted] campaign batches as well as that between the 20[redacted] campaign batches and validation batches. The drug product is manufactured from the drug substance used in the phase III clinical batches by the same method as that produced from the drug substance

taken from 20[redacted] campaign batches, and there are no differences in manufacturing scale and packaging. In addition, batch analysis has indicated great similarities in quality attributes between the drug products manufactured from these different batches of the drug substance. Based on the above, it is considered appropriate to determine the shelf life of the drug product based on the results from the long-term testing of the drug product manufactured by using phase III clinical batches for the drug substance. [redacted] drug product batches manufactured from 20[redacted] campaign batches for the drug substance have been stable for the proposed shelf-life (36 months).

PMDA accepted the applicant's response.

3. Non-clinical data

3.(i) Summary of pharmacology studies

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

No primary pharmacodynamic studies have been performed. A secondary pharmacodynamic study was conducted to investigate the effect of metreleptin versus recombinant mouse leptin using normal mice. Safety pharmacology studies were conducted to assess the effects of metreleptin on the central nervous system, cardiovascular system, respiratory system, kidney function, and gastrointestinal system.[4] No pharmacodynamic drug interaction studies have been performed.

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

Effect of subcutaneous administration of metreleptin versus recombinant mouse leptin in mice (Reference data 4.2.1.2-01)

Metreleptin (0.03, 0.3, 1, 3 mg/kg/day) or the vehicle[5] was subcutaneously administered twice daily or continuously to female mice (n = 5-8 per group) for 13 days, and clinical chemistry parameters and body composition were measured at 24 hours after the last dose (the vehicle groups were selected separately for the repeated subcutaneous administration and continuous subcutaneous administration).[6] As a result, serum glucose levels (mean ± standard error [SE]) in all dose groups (vehicle, 0.03, 0.3, 1, and 3 mg/kg/day) were 163.00 ± 9.00, 176.00 ± 12.00, 178.00 ± 19.00, 163.00 ± 8.00, and 146.00 ± 2.00 mg/dL, respectively, after repeated subcutaneous administration and 183.56 ± 14.20, 179.70 ± 11.80, 159.70 ± 6.90, 152.40 ± 7.90, and 165.00 ± 21.00 mg/dL, respectively, after continuous subcutaneous administration. Serum triglyceride levels were 53.8 ± 4.3, 85.2 ± 14.0, 60.0 ± 6.5, 58.2 ± 18.5, and 66.0 ± 10.8 mg/dL, respectively, after repeated subcutaneous administration and 70.5 ± 5.4, 69.5 ± 13.6, 64.0 ± 3.9, 69.6 ± 19.0, and 54.0 ± 9.3 mg/dL, respectively, after continuous subcutaneous administration. For body composition measured after the completion of administration, metreleptin did not cause any significant change in body weight. When compared with the vehicle groups, the body fat mass[7] decreased significantly in the ≥1 mg/kg/day groups

[4] All safety pharmacology studies were conducted as GLP-compliant studies; however, no raw data were available and the results were submitted as the reference data.

[5] Phosphate buffered saline

[6] The applicant explains that these parameters had not been analyzed statistically and additional statistical analysis cannot be performed because of unavailability of raw data.

[7] Fat was extracted from each mouse with ethyl ether and ethyl alcohol after 5-day dehydration period and then the total body fat was measured. .

after repeated subcutaneous administration and the ≥0.3 mg/kg/day groups after continuous subcutaneous administration. In addition, the body weight and food consumption[8] were measured over time. As a result, the body weight on Day 13 decreased by about 7% from baseline in the 1 mg/kg/day group after continuous subcutaneous administration[9] while there was no consistent tendency for food consumption.

Recombinant mouse leptin (0.03, 0.1, 0.3, 1 mg/kg/day) or the vehicle[5] was subcutaneously administered continuously to female mice (n = 7-9 per group) for 31 days, and clinical chemistry parameters and body composition were measured after the completion of administration. As a result, compared with the vehicle group, serum glucose levels[10], serum triglyceride levels[11], and serum total protein decreased significantly in the ≥0.3 mg/kg/day groups and serum fatty acid levels and serum cholesterol levels decreased significantly in the ≥0.1 mg/kg/day groups. For body composition measured after the completion of administration, the body fat mass decreased significantly in the ≥0.3 mg/kg/day groups compared with the vehicle group. For changes in body weight over time, the body weight on Day 31 decreased by about 13% from baseline in the 1 mg/kg/day group.[9] For changes in food consumption over time[12], compared with the vehicle group, the food consumption decreased until Day 8 and no changes were observed after Day 12.[9]

3.(i).A.(2) Safety pharmacology

3.(i).A.(2).1) Effect on the central nervous system

(a) Effect on the rat central nervous system (Reference data 4.2.1.3-01 to -04)

Metreleptin (3, 10, 30 mg/kg), the positive control[13] or the vehicle[14] was administered subcutaneously in a single dose to male rats (n = 8-10 per group[15]), and the effects of metreleptin on locomotor activity, coordinated movement, forelimb and hindlimb grip strength, and pain threshold at 0.5 hours post-dose were examined in different studies. As a result, a significant inhibition was observed, except for forelimb grip strength, in the positive control group, compared with the vehicle group. In contrast, metreleptin did not cause any effect on these parameters in all dose groups. The estimated C_{max} of metreleptin following administration of 30 mg/kg is 19,020 ng/mL[16], which is 78.9 times the maximum serum metreleptin concentration in Japanese subjects receiving the maximum clinical dose (0.08 mg/kg/day) in Japan (241 ng/mL).[17]

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[8] A 2-day total of food consumption

[9] Measurements are unknown.

[10] 214.0 ± 6.1, 219.7 ± 10.5, 193.7 ± 7.6, 175.0 ± 13.4, and 176.0 ± 10.5 mg/dL, respectively, for the vehicle group and the recombinant mouse leptin groups

[11] 81.60 ± 8.90, 86.14 ± 3.70, 67.00 ± 8.80, 55.37 ± 7.50, and 38.90 ± 3.70 mg/dL, respectively, for the vehicle group and the recombinant mouse leptin groups

[12] A 4-day total of food consumption.

[13] Morphine sulfate (20 mg/kg) was used in a study to examine the effect on pain threshold (4.2.1.3-04) and chlorpromazine hydrochloride (20 mg/kg) in other studies.

[14] Histidine solution

[15] Ten rats per group were used in a study to examine the effect on pain threshold (4.2.1.3-04) and 8 rats per group in other studies.

[16] In a pharmacokinetic study using rats (4.2.2.7-01), it was calculated to be 6 times the C_{max} of metreleptin following single subcutaneous administration of 5 mg/kg (3170 ng/mL). However, endogenous leptin is not included.

[17] The maximum serum metreleptin concentration in one Japanese patient with lipodystrophy who received multiple subcutaneous metreleptin 0.08 mg/kg/day once daily (the patient received 0.08 mg/kg/day from Weeks 8 to 20 during the 20-week study period) in the Japanese investigator-initiated trial (Study KUTR-003-1). However, endogenous leptin is included.

(b) Effect on the mouse central nervous system (Reference data 4.2.1.3-05 to -07)

Metreleptin (3, 10, 30 mg/kg), clonidine hydrochloride (positive control, 1 mg/kg), or the vehicle[14] was administered subcutaneously in a single dose to male mice (n = 6 per group), and clinical signs and behavior were observed using the Irwin test and body temperature was measured, at 0.5, 3, 6, and 24 hours post-dose. As a result, there were changes in clinical signs and behavior in the positive control group and a significant decrease in body temperature was also observed until 6 hours after administration, compared with the vehicle group. On the other hand, in the metreleptin 3 and 30 mg/kg groups, enhanced startle response was observed in 1 and 3 mice, respectively, at 0.5 hours post-dose, and in the 10 mg/kg group, a decrease in locomotor activity was observed in 2 and 3 mice, respectively, at 6 and 24 hours post-dose. Metreleptin did not cause any effect on body temperature. The applicant discusses that changes observed in the metreleptin groups are not due to the pharmacological effects of metreleptin since behavioral changes observed were mild and there were almost no changes in other relevant clinical signs and behavior.

Metreleptin (3, 10, 30 mg/kg), chlordiazepoxide hydrochloride (positive control, 20 mg/kg), or the vehicle[14] was administered subcutaneously in a single dose to male mice (n = 10 per group) and, at approximately 0.5 hours after administration, pentylenetetrazole (85 mg/kg) was administered subcutaneously to examine anticonvulsive effect. As a result, clonic convulsion was observed in 0, 10, 10, 9, and 8 mice, in the positive control group, the vehicle group, and metreleptin 3, 10, and 30 mg/kg groups, respectively. The latency to convulsion was prolonged significantly in the metreleptin 10 mg/kg group compared with the vehicle group. Tonic convulsion was observed in 0, 3, 0, 1, and 2 mice, respectively. Pentylenetetrazole (45 mg/kg) was administered subcutaneously in a similar manner and anticonvulsive effect was examined using picrotoxin (3 mg/kg) as the positive control. Although convulsion was induced in 9 mice in the positive control group, no metreleptin-induced convulsion was observed at any of the doses tested.

Metreleptin (3, 10, and 30 mg/kg), chlorpromazine hydrochloride (positive control, 4 mg/kg), or the vehicle[14] was administered subcutaneously in a single dose to male mice (n = 6 per group) and, at 0.5 hours after administration, hexobarbital (80 mg/kg) was given intraperitoneally for each group to examine the effect on hexobarbital-induced sleep. As a result, there was a significant increase in the duration of sleep in the positive control group compared with the vehicle group, but no significant changes were observed in all metreleptin groups compared with the vehicle group.

The estimated C_{max} of serum metreleptin following the administration of metreleptin (3 and 30 mg/kg) are 1101 and 4695 ng/mL, respectively[18], which are 4.6 and 19.5 times, respectively, the maximum serum metreleptin concentration in Japanese subjects receiving the maximum clinical dose (0.08 mg/kg/day) in Japan (241 ng/mL).[17]

3.(i).A.(2).2) Effect on cardiovascular system

(a) Effect on the rat cardiovascular system (Reference data 4.2.1.3-08)

[18] These estimated C_{max} values were obtained by tripling the C_{max} of metreleptin 1 and 10 mg/kg on Day 1 (367 and 1565 ng/mL, respectively) determined in the 28-day subcutaneous toxicity study in mice (4.2.3.1-01). However, endogenous leptin is not included.

The vehicle[14] or metreleptin (3, 10, 30 mg/kg) was administered subcutaneously, starting with the lowest dose, every other day, to male rats (7 rats) and the effects on blood pressure and heart rate were determined by telemetry over time up to 960 minutes post-dose[19] in each group. As a result, although there was a significant change in mean blood pressure in the metreleptin 3 and 10 mg/kg groups compared with the vehicle group (a maximum decrease of 8 mmHg in the 3 mg/kg group relative to the vehicle group), no significant changes were observed in the 30 mg/kg group relative to the vehicle group. Compared with the vehicle group, a significant decrease in heart rate was observed in the 3 and 10 mg/kg groups for up to 45 minutes after administration (a maximum decrease of 60 beats per minute in the 3 mg/kg group compared with the vehicle group), and heart rate increased significantly at 180 minutes post-dose in the 3 mg/kg group and at 720 minutes post-dose in the 30 mg/kg group compared with the vehicle group (increases of 40 and 52 beats per minute, respectively).

The applicant discusses that the increase in heart rate in the 30 mg/kg group is due to an accidental change since there was a temporary decrease in heart rate in the vehicle group at 720 minutes post-dose and it was considered to fall within the range of variations.

(b) Effect on the dog cardiovascular system (Reference data 4.2.1.3-09)

Metreleptin (5, 25 mg/kg) or the vehicle[20] was administered subcutaneously in a single dose to male dogs (n = 3 per group)[21], and blood pressure, heart rate, left ventricular pressure, maximal rate of left ventricular pressure increase, maximal rate of left ventricular pressure decrease, left ventricular pressure indicators, and pulmonary arterial pressure were determined from baseline to 24 hours post-dose, and electrocardiogram (ECG), cardiac output, stroke volume, and peripheral resistance were measured at baseline and about 1, 4, 8, 12, 18, and 24 hours post-dose. As a result, metreleptin did not have any effect on cardiovascular parameters in all dose groups.

The estimated C_{max} of serum metreleptin following the administration of metreleptin (5and 25 mg/kg) are 1689 and 5707 ng/mL, respectively, and the latter value is 23.7 times the maximum serum metreleptin concentration in Japanese subjects receiving the maximum clinical dose (0.08 mg/kg/day) in Japan (241 ng/mL).[17]

3.(i).A.(2).3) Effect on respiratory system (Reference data 4.2.1.3-05)

Metreleptin (3, 10, 30 mg/kg), clonidine hydrochloride (positive control, 1 mg/kg) or the vehicle[14] was administered subcutaneously in a single dose to male mice (n = 6 per group), and respiratory rate was determined at 0.5, 3, 6, and 24 hours post-dose. As a result, compared with the vehicle group, the respiratory rate decreased significantly in the positive control group for up to 6 hours after administration. On the other hand, although a significant decrease in respiratory rate was observed in the metreleptin 3 mg/kg group at 3

19 The analysis points included 60 minutes predose and 0, 5, 10, 15, 20, 30, 45, 60, 90, 120, 180, 240, 360, 480, 720, and 960 minutes post-dose.

20 Citrate buffer (pH 5)

21 The dose of solution administered was 5 mL/kg in the control group and the 25 mg/kg group and 1 mL/kg in the 5 mg/kg group.

hours post-dose compared with the vehicle group, metreleptin did not cause any effect in the ≥10 mg/kg groups.

The applicant discusses that the decrease in respiratory rate is not due to the pharmacological effects of metreleptin since the respiratory rates determined in the vehicle group and metreleptin 3, 10, and 30 mg/kg groups at 3 hours post-dose were 399 ± 16, 313 ± 21, 417 ± 23, and 388 ± 22 breaths per minute, respectively, and no effects were observed in the high-dose groups.

3.(i).A.(2).4) Effect on renal function (Reference data 4.2.1.3-10)

Metreleptin (3, 10, 30 mg/kg), furosemide (positive control, 20 mg/kg) or the vehicle[14] was administered subcutaneously in a single dose to male rats (n = 8 per group), and urine output, urine pH, and urine electrolyte levels (Na, K, and Cl) were determined at 0.5[22], 3, 6, and 24 hours post-dose. As a result, compared with the vehicle group, there was a significant change in these parameters in the positive control group, especially at 3 hours post-dose. On the other hand, urinary K and Cl levels increased significantly in the metreleptin 10 mg/kg group at 3 hours post-dose and the urinary Na and Cl levels decreased significantly in the 30 mg/kg group at 6 hours post-dose.

The estimated C_{max} of serum metreleptin following the administration of 3 mg/kg is 1902 ng/mL[23], which is 7.9 times the maximum serum metreleptin concentration in Japanese subjects receiving the maximum clinical dose (0.08 mg/kg/day) in Japan (241 ng/mL).[17]

3.(i).A.(2).5) Effect on gastrointestinal system

(a) Effect on gastric emptying and small intestinal transport (Reference data 4.2.1.3-11)

Metreleptin (3, 10, and 30 mg/kg), morphine sulfate (positive control, 20 mg/kg) or the vehicle[14] was administered subcutaneously in a single dose to male rats fasted overnight (n = 10 per group) and, at 30 minutes after administration, 1 mL of carbon dust[24] was given orally for each group. At 30 minutes after the administration, the stomach and small intestine were removed and the gastric emptying and small intestinal transport were evaluated. As a result, compared with the vehicle group, a significant inhibition was observed in the positive control group. In contrast, metreleptin did not cause any effect in all dose groups.

(b) Effect on the motility of the isolated ileum (Reference data 4.2.1.3-12)

Following the application of metreleptin (0.5, 5, 50 μg/mL) or the vehicle[14], to ileum isolated from male guinea pigs (n = 4 per constrictor group), starting with the lowest dose[25], the effect on the contraction elicited

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22 Since the urine output was low in the metreleptin groups and the vehicle group, urine pH and urine electrolyte levels were not measured in some rats and these parameters were not analyzed statistically.

23 The estimated C_{max} value was calculated to be third-fifths of the C_{max} of serum metreleptin following single subcutaneous administration of 5 mg/kg (3170 ng/mL) in a pharmacokinetic study using rats (4.2.2.7-01). However, endogenous leptin is not included.

24 It contains 10% carbon dust and 30% flour.

25 After two sections were isolated from each guinea pig, metreleptin or the vehicle was applied to each of them. Metreleptin was applied by dose titration to each of the constrictor groups.

by acetylcholine (100 nmol/L), histamine (100 nmol/L), serotonin (100 nmol/L), and barium chloride (300 μmol/L)[26] was examined. As a result, metreleptin did not cause any effect.

In addition, metreleptin 50 μg/mL is 207 times the maximum serum metreleptin concentration in Japanese subjects receiving the maximum clinical dose (0.08 mg/kg/day) in Japan (241 ng/mL).[17]

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

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

Since no primary pharmacodynamic studies of metreleptin had been performed, PMDA asked the applicant to explain the reasons for not conducting the studies, by examining the mechanism of action of metreleptin and its *in vivo* efficacy based on the information such as *in vivo* distribution of leptin receptors, differences in bioactivity between endogenous leptin and metreleptin, and characteristics of patients with lipodystrophy and disease-model animals.

The applicant responded as follows:

The results from the comparison of sequence homology of endogenous leptin and leptin receptors between humans and several species of animals indicated that the homology of endogenous leptin in mice, rats, and dogs was 83%, 82%, and 81%, respectively, and that of leptin receptors was 75%, 75%, and 83%, respectively. A functional receptor, Ob-Rb, which is one of the leptin receptors (obesity receptor [Ob-R]) with intracellular signaling domains, is expressed in the hypothalamus, kidneys, lungs, adrenal glands, blood cells including immune cells, etc.[27] In addition to metreleptin's effects on appetite suppression and body weight regulation via hypothalamus, leptin is also known to have an effect on hormone secretion from the diencephalon-hypothalamo-hypophyseal system, the nervous system (e.g., increased blood pressure), the immune system, osteogenesis, etc. Metabolic regulation, a major effect of metreleptin, has been reported to be common to all species.[28]

No *in vitro* studies have been performed to directly compare the activity of metreleptin with that of human endogenous leptin. However, a study on intracerebroventricular administration of recombinant human leptin in ob/ob mice (Imagawa K, et al. *J Biol Chem.* 1998;273:35245-9) revealed that there was a decrease in food consumption and body weight at doses that did not differ greatly from those used in an intracerebroventricular administration study of recombinant mouse leptin (Campfield LA, et al. *Science.* 1995;269:546-9), which infers that the effects of metreleptin and human endogenous leptin on the organs where leptin receptors are present are similar. When metreleptin or recombinant mouse leptin was administered to normal mice in the secondary pharmacology study (4.2.1.2-01), administration of recombinant mouse leptin reduced the serum glucose and triglyceride levels, and both metreleptin and recombinant mouse leptin showed a similar tendency for the effect on body weight, etc.

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[26] They are acetylcholine hydrochloride, histamine dihydrochloride, 5-hydroxytryptamine creatinine sulfate, and barium chloride, respectively.

[27] Vaisse C, et al. *Nature Genet.* 1996;14:95-7, Hoggard N, et al. *Biochem Biophys Res Commun.* 1997;232:383-7, Cioffi JA, et al. *Nature Med.* 1996;2:585-9

[28] Agata J, et al. *Am J Hypertens.* 1997;10:1171-4, Flier JS, et al. *Proc Natl Acad Sci USA.* 1997;94:4242-5, Lord GM, et al. *Nature.* 1998;394:897-901, Khosla S, et al. *Endocrinology.* 2002;143:4161-4, Wylie ARG. *Animal.* 2011;5:246-67

As an animal model of lipodystrophy, transgenic mice (aP2-nSREBP-1cTg mice and A-ZIP mice) lacking adipocytes due to the inhibition of adipocyte differentiation (maturation) are available. Although the mechanism of loss of adipocytes is speculated to be different from that in human lipodystrophy principally caused by various genetic mutations and autoimmune disease, decreased blood leptin concentrations, hepatic steatosis, dyslipidemia, increased blood insulin concentration and blood glucose level, insulin resistance, etc., were observed in these animal models, expressing phenotypes similar to those observed in lipodystrophy. There are no results from non-clinical studies examining the effect of metreleptin in these animal models nor published articles. However, following 12-day continuous subcutaneous administration of recombinant mouse leptin 5 μg/day to aP2-nSREBP-1c Tg mice, plasma insulin concentrations, blood glucose levels, triglyceride levels in the liver, histology of hepatic steatosis, etc., have been reported to improve to the similar level observed in the wild-type mice (Shimomura I, et al. *Nature.* 1999;401:73-6). When mice are prepared by mating A-ZIP mice with Tg mice which express high levels of mouse leptin, an increase in serum mouse leptin concentrations, decreases in blood glucose, plasma triglyceride, free fatty acid, and insulin levels, and improvements in insulin resistance and histology of hepatic steatosis have been reported in the F1 mouse (Ebihara K, et al. *Diabetes.* 2001;30:1440-8). Neither report provides data on HbA1c as the indicator of long-term blood glucose control and AST and ALT as the indicators for liver impairment. However, based on the fact that the normalization of blood glucose levels and the improvement of blood lipid levels and hepatic steatosis are achieved by administration of recombinant mouse leptin to a mouse model of lipodystrophy, an improvement in these indicators is also expected and the treatment effect of metreleptin in patients with lipodystrophy could be expected.

Based on the above, although no primary pharmacodynamic studies of metreleptin have been conducted, the mechanism of action and efficacy of metreleptin is considered to have been demonstrated because (a) there are publications on the physiological action of endogenous leptin and the efficacy of leptin replacement in an animal model of lipodystrophy and (b) results from a foreign NIH clinical study started in 2000 (Study [redacted]1265/20[redacted]0769) and a Japanese clinical research started in 2002 (Study KUTR-003-0) have shown improvement in glucose metabolism disorders and lipid metabolism disorders in patients with lipodystrophy. Therefore, no primary pharmacodynamic studies were performed.

Since there is the possibility that unphysiologically high serum leptin concentrations in blood could have occurred in the secondary pharmacology study using normal mice (4.2.1.2-01) and serum leptin concentrations exceeding the mean concentration in Japanese healthy adult subjects were observed in some subjects in the Japanese investigator-initiated trial (Study KUTR-003-1) and Japanese clinical research (Study KUTR-003-0), PMDA asked the applicant to explain whether or not excessively-stimulated leptin receptors affect efficacy.

The applicant responded as follows:
Although a leptin-induced decrease in expression of leptin receptors has been reported for cultured cells (Hikita M, et al. *Biochem Biophys Res Commun.* 2000;271:703-9), there have been no reports of decreased *in*

vivo expression of leptin receptors at present. Since leptin signaling is subject to negative feedback loop (Bjorbak C, et al. *J Biol Chem.* 2000; 275:40649-57), it is inferred that the effect of leptin is regulated to maintain a consistent level within an organism. Although some patients had unphysiologically high serum leptin concentrations in the Japanese investigator-initiated trial (Study KUTR-003-1) and Japanese clinical research (Study KUTR-003-0), the efficacy of metreleptin was not reduced. Thus, the applicant considers that excessively-stimulated leptin receptors are unlikely to affect efficacy at present.

PMDA considers as follows:
Non-clinical evaluation is not adequate, but considering the explanations on endogenous leptin and disease-model animals, and the rareness and seriousness of the target disease, the applicant's response is acceptable. However, it is necessary to continue to evaluate the efficacy of metreleptin in the clinical data section [for efficacy in humans, see "4.(iii).B.(2) Efficacy"].

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

PMDA asked the applicant to explain the safety of metreleptin in humans based on the results of safety pharmacology studies.

The applicant responded as follows:
With respect to the effect on the central nervous system, anticonvulsive effect was observed in the study to examine the effect on the mouse central nervous system (4.2.1.3-06) and the serum metreleptin concentration after the maximum dose at which no anticonvulsive effect had been observed was 4.6 times that in human subjects receiving the maximum clinical dose (4.2.1.3-06). The anticonvulsive effect observed following metreleptin administration is considered to have been partly attributed to the suppression of neuronal excitation by metreleptin-induced activation of janus kinase 2 (JAK2)/phosphatidylinositol 3 kinase (PI3K) signal transduction pathways and inhibition of synaptic transmission based on the following reasons: (a) recombinant mouse leptin inhibits α-amino-3-hydroxy-5-methyl-4-isoxazole propionate (AMPA) receptor-mediated synaptic transmission, (b) JAK2/PI3K signal transduction pathways is suggested to be involved in the inhibition of synaptic transmission (Xu L, et al. *J Clin Invest.* 2008;118[1]:272-280), and (c) ob/ob mice have higher sensitivity to pentylenetetrazol-induced seizures compared with the wild-type mice (Erbayat-Altay E, et al. *Neurosci Lett.* 2008;433:82-6). However, metreleptin is unlikely to have a serious impact on the central nervous system at present based on the following finding: headache, which was an adverse event reported most frequently as the one falling under the category of "Nervous system disorders" in the MedDRA system organ class, occurred in 2 of 4 subjects (50.0%), 7 of 11 subjects (63.6%), and 0 subjects (0%), respectively, in the Japanese investigator-initiated trial (Study KUTR-003-1), Japanese clinical research (Study KUTR-003-0), and Japanese investigational study (Study KUTR-003-2), but all episodes of headache were mild in severity and the outcome was "resolved" in most cases.

The effect on the cardiovascular system was assessed. Metreleptin is very unlikely to affect the cardiovascular system by inhibiting the hERG channel intracellularly by membrane transport since metreleptin is a polypeptide consisting of 147 amino acid residues with a molecular weight of about 16 kDa.

Also, subcutaneous metreleptin did not affect ECG at doses up to 25 mg/kg in the study to examine the effect on the dog cardiovascular system (4.2.1.3-09), and metreleptin treatment did not cause any change in ECG data in the 1-, 3-, and 6-month subcutaneous toxicity study in dogs (4.2.3.2-02). These findings suggest that metreleptin is unlikely to have an effect on the cardiovascular system. The effect on the respiratory system was assessed. Although only the effect on respiratory rate had been examined in the study to examine the effect on the mouse respiratory system (4.2.1.3-05), subcutaneous metreleptin did not affect breathing at doses up to 30 mg/kg. In the study to examine the effect on the dog cardiovascular system (4.2.1.3-09) and 28-day subcutaneous toxicity study in mice (4.2.3.1-01), metreleptin also did not cause any change in observations of clinical signs. The results suggest that metreleptin does not have a serious impact on the respiratory system.

PMDA accepts the applicant's response, but considers that it is necessary to continue to examine the safety in humans in Clinical Section [for safety in humans, see "4.(ii).B.(1) Effect of unphysiologically high serum leptin concentrations on safety" and "4.(iii).B.(3) Safety"].

3.(ii) Summary of pharmacokinetic studies

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

Pharmacokinetics were determined after single intravenous or subcutaneous administration of metreleptin in mice and dogs. The pharmacokinetics of metreleptin following repeated subcutaneous administration in mice and dogs were also determined. The concentrations of mouse and rat serum metreleptin were measured by enzyme immunoassay and the lower limits of quantification were 0.088 and 0.5 ng/mL, respectively. The concentrations of dog serum metreleptin were measured by electrochemiluminescence immunoassay and the lower limit of quantification was 3.0 ng/mL.

3.(ii).A.(1) Absorption (4.2.2.2-01 to -04)

The pharmacokinetic parameters of metreleptin following single intravenous or subcutaneous administration in male mice (N = 3/timepoint/group) and male dogs (n = 4 per group) were as shown in Table 4.

Table 4. Pharmacokinetic parameters of metreleptin following single intravenous or subcutaneous administration

Animal species	Route	Dose (mg/kg)	N	t_{max} (h)	C_{max} (ng/mL)	AUC_{inf} (ng·h/mL)	CL_{tot} (mL/h/kg)	$V_{d,ss}$ (mL/kg)	BA (%)	$t_{1/2}$ (h)		
										α phase	β phase	γ phase
Mice	i.v.[a)]	0.3	42	-	-	491	611	142	-	0.0886	0.446	NC
		1	42	-	-	1470	681	146	-	0.0834	0.491	NC
		3	42	-	-	4530	663	171	-	0.0904	0.484	7.18
		10	42	-	-	15,800	633	158	-	0.0558	0.476	8.86
	s.c.[b)]	0.3	39	0.28	377	348	-	-	70.8	0.408	NC	-
		1	39	0.14	1520	1230	-	-	83.7	0.388	NC	-
		3	39	0.50	3810	3780	-	-	83.4	0.379	4.541	-
		10	39	0.39	12,600	14,000	-	-	88.7	0.436	8.626	-
Dogs	i.v.	0.3	4	-	-	1450	215	193	-	0.16	1.16	-
		3	4	-	-	16,400	194	165	-	0.19	1.46	-
	s.c.	0.3	4	2.8	180	1320	-	-	91	-	-	2.13
		3	4	4.0	1080	11,700	-	-	72	-	-	2.66

Mean; -, not applicable; NC, not calculated;
t_{max}, time to reach maximum serum concentration; C_{max}, maximum serum concentration; $AUC_{inf:}$, area under the serum concentration-time curve extrapolated to infinity; CL_{tot}, total clearance; $V_{d,ss}$, steady-state volume of distribution; BA, bioavailability; $t_{1/2}$, elimination half-life
a) For 3 rats/timepoint, blood samples were collected at 14 timepoints to calculate the parameters.
b) For 3 rats/timepoint, blood samples were collected at 13 timepoints to calculate the parameters.

The pharmacokinetic parameters of metreleptin following 14-day repeated subcutaneous administration in male mice (n = 3/timepoint/group) and male and female dogs (n = 2/sex/group) were as shown in Table 5.

Table 5. Pharmacokinetic parameters of metreleptin following 14-day repeated subcutaneous administration

Animal species	Dose (mg/kg)	N (male/female)	Date of measurement	t_{max} (h)	C_{max} (ng/mL)	AUC_{0-24h} (ng·h/mL)
Mice[a)]	1	45/0	Day 1	0.167	1440	1263
		45/0	Day 7	0.28	1393	1453
		45/0	Day 14	0.39	1534	1891
	10	45/0	Day 1	0.39	10,700	10,256
		45/0	Day 7	0.50	12,480	12,644
		45/0	Day 14	0.28	14,700	17,937
Dogs	0.3	2/2	Day 1	4.0	155	1220
		2/2	Day 7	2.5	194	1240
		2/2	Day 14	2.0	361	2520
	3	2/2	Day 1	4.0	1440	11,500
		2/2	Day 7	2.5	2180	13,500
		2/2	Day 14	2.3	2750	21,600

Mean; t_{max}, time to reach maximum serum concentration; C_{max}, maximum serum concentration; AUC_{0-24h}, area under the serum concentration-time curve from 0 to 24 hours
a) For 3 rats/timepoint, blood samples were collected at 15 timepoints to calculate the parameters.

In male mice, the ratios of C_{max} on Days 7 and 14 to Day 1 were 0.97 and 1.07, respectively, in the 1 mg/kg group and 1.17 and 1.37, respectively, in the 10 mg/kg group; and the ratios for AUC_{0-24h} were 1.15 and 1.5, respectively, in the 1 mg/kg group and 1.23 and 1.75, respectively, in the 10 mg/kg group. In male and female dogs, the ratios for C_{max} were 1.25 and 2.33, respectively, in the 0.3 mg/kg group, 1.51 and 1.91, respectively, in the 3 mg/kg group; the ratios for AUC_{0-24h} were 1.01 and 2.14, respectively, in the 0.3 mg/kg group and 1.19 and 1.92, respectively, in the 3 mg/kg group.

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

No distribution studies have been performed.

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

No metabolism studies have been performed.

3.(ii).A.(4) Excretion (4.2.2.5-01)

The pharmacokinetic parameters of metreleptin 10 mg/kg following single intravenous administration in control mice, sham-operated mice, and mice with bilateral nephrectomy (male mice, n = 3/timepoint/group) were as shown in Table 6.

Table 6. Pharmacokinetic parameters of metreleptin following single intravenous administration

Treatment group[a)]	N	AUC_{inf} (ng·h/mL)	CL_{tot} (mL/h/kg)	$V_{d,ss}$ (mL/kg)
Control mice	42	14,400	697	203
Sham-operated mice	42	15,100	662	195
Mice with bilateral nephrectomy	42	439,000	22.8	125

AUC_{inf}, area under the serum concentration-time curve extrapolated to infinity; CL_{tot}, total clearance; $V_{d,ss}$, steady-state volume of distribution
a) For 3 mice/timepoint, blood samples were collected at 14 timepoints to calculate the parameters.

The serum leptin concentrations (mean ± standard deviation [SD]) at 24 hours post-dose were 0.70 ± 0.44, 0.65 ± 0.40, and 1333 ± 205 ng/mL, in control mice, sham-operated mice, and mice with bilateral nephrectomy, respectively.

3.(ii).A.(5) Equivalence of the drug products (4.2.2.7-01)

The pharmacokinetic parameters of metreleptin following single subcutaneous administration of lyophilized formulation 5 mg/kg or frozen formulation[3] 5 mg/kg in male rats (16 rats) using a 2-phase crossover design were as shown in Table 7. The washout period was 2 days for each treatment period.

Table 7. Pharmacokinetic parameters of metreleptin following single subcutaneous administration of lyophilized formulation or frozen formulation

Drug product	N	t_{max} (h)	C_{max} (ng/mL)	$AUC_{0\text{-}24h}$ (ng·h/mL)
Lyophilized formulation	16	0.833 ± 0.408	3170 ± 361	6450 ± 664
Frozen formulation	16	0.846 ± 0.427	3290 ± 626	6820 ± 485

Mean ± SD
t_{max}, time to reach maximum serum concentration; C_{max}, maximum serum concentration; $AUC_{0\text{-}24h}$, area under the serum concentration-time curve from 0 to 24 hours

The C_{max} and $AUC_{0\text{-}24h}$ ratios of lyophilized formulation to frozen formulation (lyophilized formulation treatment/frozen formulation treatment) and their 90% confidence intervals (CIs) were 99.4 (95.4, 101.9) and 95.9 (97.1, 98.8), respectively.

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

Pharmacokinetics in patients with renal impairment

The applicant discussed excretion of metreleptin as follows:

When mice were administered a single intravenous dose of metreleptin 0.3 to 10 mg/kg (4.2.2.2-01), the total clearance (CL_{tot}) ranged from 611 to 681 mL/h/kg and it was similar to the glomerular filtration rate in mice (595 mL/h/kg; Takahashi N, et al. *Kidney Int.* 2007;71:266-71). When dogs were administered a single intravenous dose of metreleptin 0.3 to 3 mg/kg (4.2.2.2-03), the CL_{tot} ranged from 194 to 215 mL/h/kg, which was approximately 85% of the glomerular filtration rate in dogs (246 mL/h/kg; Finco DR, et al. *Am J Vet Res.* 1981;42:1874-7). When mice with bilateral nephrectomy were administered a single intravenous

dose of metreleptin 10 mg/kg (4.2.2.5-01), the CL_{tot} was approximately 3% of that in control mice and sham-operated mice, hence, metreleptin was eliminated primarily from the body of mice by the kidney (>95%). The above results indicate that metreleptin is eliminated primarily by renal excretion through glomerular filtration.

Based on considerations of the results from non-clinical pharmacokinetic studies, PMDA asked the applicant to explain the elimination routes for metreleptin in humans.

The applicant responded as follows:
When human subjects were administered multiple intravenous doses of metreleptin 0.3 to 3 mg/kg (Study LEPT-[redacted]0121), the total clearance (CL) on Day 1 ranged from 79.6 to 95.8 mL/h/kg, which is similar to the glomerular filtration rate in humans (120 mL/min/70 kg = 103 mL/h/kg; Rowland M, et al. *Clinical Pharmacokinetics Concepts and Applications. 3rd ed*. ed. by Rowland M, et al. Lippincott Williams & Wilkins, Philadelphia, 1995;171). The maximum renal excretion percentage calculated from arterial endogenous leptin concentrations and endogenous leptin concentrations in the renal vein in healthy adult subjects has been reported to be about 80% (Meyer C, et al. *Am. J. Physiol Endocrinol Metab.* 1997;273:E903-7.). The above results indicate that as in mice and dogs, metreleptin is eliminated primarily by renal excretion through glomerular filtration in humans.

PMDA asked the applicant to explain the necessity of exercising caution for patients with renal impairment.

The applicant responded as follows:
When mice with bilateral nephrectomy were administered a single intravenous dose of metreleptin 10 mg/kg (4.2.2.5-01), serum leptin concentrations were increased and the maximum renal excretion rate is estimated to be about 80% in humans (Meyer C, et al. *Am. J. Physiol Endocrinol Metab.* 1997;273:E903-7). Therefore, metreleptin is excreted primarily by the kidney. Since no clinical studies in patients with renal impairment have been conducted, the effect of renal impairment on pharmacokinetics is unknown but renal impairment may result in an increase in serum leptin concentrations. Thus, it is considered necessary to exercise caution for patients with renal impairment and a caution statement will be included in the Important Precautions section of the package insert.

PMDA accepts the explanation from the perspective of pharmacokinetics, but considers that it is necessary to continue to examine the necessity of including the caution statement in the package insert in Clinical Section [“4.(iii).B.(6).3) Patients with renal impairment and elderly patients”].

3.(iii) Summary of toxicology studies

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

Toxicity studies of metreleptin conducted include repeat-dose toxicity, genotoxicity, reproductive and developmental toxicity, a local tolerance study, and other toxicity studies (an antigenicity study and a blood

compatibility study). No single-dose toxicity studies were performed, but single-dose toxicity was evaluated in a 28-day subcutaneous toxicity study in mice.

3.(iii).A.(1) Repeat-dose toxicity

Repeated subcutaneous dose toxicity studies in mice (28 days and 3 and 6 months), dogs (3 weeks, 28 days, and 1, 3 and 6 months), and male rats (14 days) and repeated intravenous dose toxicity studies in mice (28 days) and dogs (4 weeks) were performed. The major toxicity findings included gastric mucosal erosion and mixed cell infiltration into the perirenal fat in mice, and hemorrhage (sclera, gingiva, stomach, intestine, and bladder) and perivasculitis (injection site, adipose tissue, kidney, liver, etc.) in dogs. Since antibody production occurred at 1 month post-dose in dogs and it was considered to affect the measurement system for serum metreleptin concentrations, only the extent of exposure on Days 1, 7, and 14 was used for evaluation.[29]

3.(iii).A.(1).1) Mouse 28-day subcutaneous toxicity study (4.2.3.1-01)

In this toxicity study, the vehicle[30] or metreleptin (1, 10, 100 mg/kg/day) was administered subcutaneously once daily to male and female mice (Swiss albino, n = 35/sex/dose) for 28 days. Deaths occurred in 2 male and 2 female mice in the 100 mg/kg/day group on Day 9 or 10, and inhibition of body weight gain, a decrease in food consumption, changes in blood chemistry parameters such as a decrease in triglycerides, and a decrease in adipose tissue were observed in the metreleptin groups, indicating changes caused by the pharmacological action of metreleptin. Cellulitis at the injection site was observed in the metreleptin groups, and decreases in thymus weight and spleen weight, centrilobular hepatocyte degeneration, lymphocytolysis in lymphoid tissues (thymus, spleen, mesenteric lymph nodes, etc.), gastric mucosal erosion, and a decrease in zymogen granules in the pancreas were observed in the ≥10 mg/kg/day groups. Since there were no deaths or serious changes in clinical signs at 1 week after the start of treatment, the approximate lethal dose for mice was determined to be >100 mg/kg/day.

3.(iii).A.(1).2) Mouse 3- and 6-month subcutaneous toxicity study (4.2.3.2-01)

In this toxicity study, the vehicle[30] or metreleptin (0.3, 1, 3, 10, 30 mg/kg) was administered subcutaneously once daily to male and female mice (Swiss albino, n = 45/sex/dose) for 3 or 6 months (including 28-day recovery assessment after 6-month treatment). Deaths occurred in a total of 10 mice (2 female and 2 male mice in the 3 mg/kg/day group, 2 female mice in the 10 mg/kg/day group, and 2 male and 2 female mice in the 30 mg/kg/day group). Since 9 mice, excluding 1 female mouse in the 30 mg/kg/day (died on Day 33), died on Day 91, fasting and water deprivation before necropsy were considered more likely to cause excessive exacerbation of conditions.[31] Examinations at the end of the 3-month treatment revealed decreased skin temperature, hunchback position, dehydration, decreased locomotor activity, inhibition of body weight gain and decrease in food consumption, decreases in liver, spleen, heart, and kidney weight, darkening of gastrointestinal contents, darkening of the stomach and reduced thymic size, decreased mesenteric and

[29] The measurement of plasma metreleptin concentrations and antibodies in mice was performed in the 28-day repeated subcutaneous dose toxicity study (4.2.3.1-01), and antibody production was confirmed at 28 days post-dose.

[30] 5% sorbitol containing 10 mmol/L histidine

[31] In the 6-month treatment assessment group and recovery assessment group, overnight fasting and water deprivation performed in the 3-month treatment assessment group were not implemented before planned autopsy.

perirenal adipose tissue, lymphocytolysis in the spleen, lymph nodes, and thymus, a decrease in zymogen granules in the pancreas, and cellulitis at the injection site (increased incidence) in the metreleptin groups. Increases in urea nitrogen, decreased glucose, cholesterol, and triglyceride, centrilobular hepatocyte degeneration, and gastric mucosal erosion were observed in the ≥1 mg/kg/day groups. Mixed cell infiltration into the perirenal fat in the ≥3 mg/kg/day groups, a decrease in hemoglobin levels in the ≥10 mg/kg/day groups, decreases in red blood cell count and hematocrit levels in the 30 mg/kg/day group. The mixed cell infiltration into perirenal fat appeared to be a change due to continuous induction of macrophages associated with lipolysis, the pharmacological action of metreleptin.[32] Since the inhibition of food consumption is known to result in stress-induced gastric mucosal erosion (Levin S, et al. *Toxicologic Pathology.* 1993;21:1-14), the gastric mucosal erosion observed in this study is considered to be a stress-induced change due to feeding suppression. Examinations at the end of 6-month treatment revealed no changes in clinical signs, but indicated inhibition of body weight gain and a decreasing trend of food consumption in the metreleptin groups. The findings from the blood chemistry and histopathological examinations were similar to those obtained at the end of the 3-month treatment, but their severity and incidence tended to decrease. All findings were recoverable. Based on the above, the no observed adverse effect level (NOAEL) was determined to be 1 mg/kg/day in this study.

3.(iii).A.(1).3) Dog 1-, 3-, and 6-month subcutaneous toxicity study (4.2.3.2-02)

In this toxicity study, the vehicle[30] or metreleptin (0.05, 0.15, 0.5, 1.5, 5 mg/kg) was administered subcutaneously once daily to male and female dogs (beagle; control group, n = 12/sex/dose; metreleptin 0.05, 0.15 mg/kg/day groups, n = 9/sex/dose; metreleptin 0.5, 1.5, 5 mg/kg/day groups, n = 14/sex/dose) for 1, 3, or 6 months (including the group with 1-month administration of doses ≥0.5 mg/kg/day followed by a washout period of 1 month [first recovery group, n = 2/sex], the group with a washout period of 4 or 5 months after Week 8 of treatment with 5 mg/kg/day [unscheduled recovery group, n = 1/sex], the group with a washout period of 3 or 4 months after 3-month administration of 5 mg/kg/day [second recovery group[33], n = 5/sex], and the group with a washout period of 1 month after 6-month administration of the vehicle or 0.5 and 1.5 mg/kg/day [third recovery group, n = 3/sex]). No deaths occurred in all groups and 1 female mouse in the 0.5 mg/kg/day group and 1 male mouse in the 1.5 mg/kg/day group were dying in Week 6 and Week 8, respectively, due to severe weight loss. Examinations at the end of the 1-month treatment revealed weight loss, a decrease in food consumption, and a decrease in thymus weight in the metreleptin groups, emaciation and relaxation of skin in the ≥0.15 mg/kg/day groups, decreased white blood cell count and increased cortisol in the ≥0.5 mg/kg/day groups, and visible ribs, local gingival hemorrhage, prolonged capillary refilling time, delayed papillary reflex, decreases in albumin, total protein, cholesterol, etc., thymic atrophy, and hypertrophy of thyroid follicular epithelial cells in the ≥1.5 mg/kg/day groups; inanimation, pupillary dilatation, ulcers on the gastric mucosal surface, hemorrhage (sclera, gastric submucosa, duodenal mucosal surface, bladder submucosa, etc.), purulent inflammation (stomach, ileum, cecum, colon, and rectum), and

[32] Kosteli A, et al. *J Clinical Investigation.* 2010;120:3466-79, Friedman JM, et al. *Nature.* 1998;395:763-70

[33] The applicant considered that dogs in the 5 mg/kg/day group cannot tolerate 6-month treatment due to the worsening of clinical signs before the completion of 3-month administration. Among 6 male and 6 female dogs including the test animals evaluated for 6-month toxicity and the recovery group, treatment was discontinued in 1 male and 1 female after Week 8 for 4 or 5 months and they were included in the unscheduled recovery group. Treatment was discontinued in the remaining 5 male and 5 female dogs for 3 or 4 months after the completion of 3-month administration and they were included in the second recovery group.

perivasculitis (adipose tissue, liver, kidney, etc.) were observed in the 5 mg/kg/day group. Although decreases in thyroid-stimulating hormone and total thyroxine were observed in the first recovery group, other findings showed a recovery trend. Examinations at the end of the 3-month treatment revealed a decrease in adipose tissue and increases in the incidence and severity of perivasculitis at the injection site in the metreleptin groups, weight loss, a decrease in food consumption, and local gingival hemorrhage in the ≥0.15 mg/kg/day groups, decreases in red blood cell count, hemoglobin levels, and hematocrit levels, red gastric and intestinal contents, and reduced thymic size in the ≥0.5 mg/kg/day groups, decreased skin temperature, decreases in total iron binding capacity, etc., and increased globulin in the ≥1.5 mg/kg/day groups, and decreased white blood cell count and follicular atrophy of spleen in the 5 mg/kg/day group. All findings were reversible in the unscheduled recovery group and second recovery group.[33] Evaluations at the end of the 6-month treatment revealed weight loss, a decrease in food consumption, an increasing trend of cortisol, and others in the ≥0.15 mg/kg/day groups. Although cortisol and urea nitrogen had continued to increase in the 1.5 mg/kg group of the third recovery group, other findings were reversible. Based on the above, the NOAEL was determined to be 1.5 mg/kg/day in this study.

3.(iii).A.(1).4) Mouse 28-day intravenous toxicity study (4.2.3.2-03)

In this toxicity study, the vehicle[34] or metreleptin (1, 10, 100 mg/kg) was administered intravenously once daily to male and female mice (CD-1, n = 25/sex/dose) for 28 days. In the metreleptin groups, loss of adipose tissue was observed, inhibition of body weight gain and a decrease in food consumption were observed in the ≥10 mg/kg/day groups. In the 100 mg/kg/day group, deaths occurred in 3 male mice and 2 female mice, and decreases in parameters such as albumin, total protein, and cholesterol, increases in urea nitrogen and corticosterone, renal tubular degeneration, inflammation of the renal pelvis, deposition of protein-like substances, and hydronephrosis were observed. Based on the above, the NOAEL was determined to be 10 mg/kg/day under the conditions of this study.

3.(iii).A.(1).5) Male mouse 28-day intravenous toxicity study (4.2.3.2-04)

In this toxicity study, the vehicle[34] or metreleptin (10, 30, 60, 90, 100 mg/kg) was administered intravenously once daily to male mice (CD-1, n = 10 per group) for 28 days. There were no deaths. Inhibition of body weight gain and a decrease in food consumption were observed in the metreleptin groups, a decrease in triglyceride levels and loss of adipose tissue were observed in the ≥30 mg/kg/day groups, hydronephrosis was noted in the ≥60 mg/kg/day groups, and inflammation of the renal pelvis and deposition of protein-like substances were observed in the ≥90 mg/kg/day groups. Based on the above, the NOAEL was determined to be 30 mg/kg/day under the conditions of this study.

3.(iii).A.(1).6) Rat 14-day subcutaneous toxicity study (4.2.3.2-05)

In this toxicity study, the vehicle[35], metreleptin acetate[36], or lyophilized formulation of metreleptin 5 mg/kg (5, 20, 50 mg/mL)[37] was administered subcutaneously once daily to male rats (SD, n = 6 per group) for 14

[34] 5% sorbitol containing 10 mmol/L sodium acetate

[35] 0.017% Tween 20 containing 2.44% glycine

[36] The active ingredient of metreleptin is not acetate.

days (including 2-week washout group [n = 2 per group]). In the metreleptin groups, there were no deaths but inhibition of body weight gain, a decrease in food consumption, and a decrease in insulin were observed. Increases in urea nitrogen and urea nitrogen/creatinine ratio were observed in the metreleptin acetate 20 mg/mL/day and 50 mg/mL/day groups and an increase in alkaline phosphatase, etc. in the metreleptin acetate 20 mg/mL/day group. Concentration-dependent, non-purulent inflammation at the injection site was observed in all groups. All findings were reversible in the washout group. Based on the above, no major difference in effect was considered to be observed between metreleptin acetate and lyophilized formulation.

3.(iii).A.(1).7) Dog 28-day subcutaneous toxicity study (4.2.3.2-06)

In this toxicity study, the vehicle[34] or metreleptin (0.5, 5 mg/kg) was administered by subcutaneous bolus injection or continuous infusion[38] (0.01 mL/kg/h) to male and female dogs (beagle, n = 3/sex/dose) for 28 days. Regardless of the route of administration, dehydration, emaciation, a decrease in food consumption, weight loss, decreases in triglycerides, etc., a decrease in organ weight (heart, spleen, thymus, etc.), decreased mesenteric and perirenal adipose tissue, a decrease in bone marrow cells, atrophy of lymphoid tissue in the thymus, and a decrease in zymogen granules in the pancreas were observed in the metreleptin groups.

3.(iii).A.(1).8) Dog 3-week subcutaneous toxicity study (4.2.3.2-07)

In this toxicity study, the vehicle[39], metreleptin, or pegylated metreleptin (1.5, 3.5 mg/kg/dose [three times weekly] or 1.5 and 5.25 mg/kg/dose [two times weekly]) was administered subcutaneously to male and female dogs (beagle, n = 2/sex/dose) for 3 weeks. As the findings common to the metreleptin groups and pegylated metreleptin groups, emaciation and decreases in adipose tissue, insulin, etc. were observed in all groups and decreases in red blood cell count, hemoglobin levels, and hematocrit levels were observed in the 5.25 mg/kg/dose group. In the pegylated metreleptin groups, decreases in total protein and cholesterol, increases in urea nitrogen and urea nitrogen/creatinine ratio, dark red gastric and gastrointestinal contents, and proximal tubular vacuolization were observed in the ≥1.5 mg/kg/dose groups, and tremor and an increase in creatine kinase were noted in the 5.25 mg/kg/dose group. Based on the above, the pegylated metreleptin group was considered to show a marked change compared with the metreleptin group.

3.(iii).A.(1).9) Dog 4-week intravenous toxicity study (4.2.3.2-08)

In this toxicity study, the vehicle[34] or metreleptin (0.5, 1.5, 5 mg/kg) was administered intravenously once daily to male and female dogs (beagle, n = 3/sex/dose) for 4 weeks. In the metreleptin groups, weight loss, a decrease in food consumption, a decrease in organ weight (liver, kidney, heart, etc.), a decrease in adipose tissue, and loss of lymphocytes in the thymus were observed, and in the ≥1.5 mg/kg/day groups, atony of the skin, decreased bowel movements, emaciation, transient inanimation, shivering and vocalization, decreased triglycerides and increases in urea nitrogen, etc., edema in the gastric submucosa, and loss of bone-marrow

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37 50 mg/mL is only available for a lyophilized formulation.

38 The solution administered was 0.25 mL/kg/day (0.5 mg/kg group, 2 mg/mL; 5 mg/kg group, 20 mg/mL).

39 Phosphate buffered saline

hematopoietic cells and feeder cells were noted. Based on the above, the NOAEL was determined to be 5 mg/kg/day under the conditions of this study.

3.(iii).A.(2) Genotoxicity (4.2.3.3-01 to -04)

A bacterial reverse mutation assay, a gene mutation assay using cultured Chinese hamster ovarian cells (CHO cells), a chromosomal aberration assay using CHO cells, and a mouse bone marrow micronucleus assay were performed. In the chromosomal aberration assay, the frequency of structural chromosomal aberrations increased by 9% when treated at 2000 μg/mL in the presence of metabolic activation for 6 hours and by 11% when continuously treated at 2000 μg/mL for 44 hours; however, reproducibility was not confirmed in a retest. As stated above, metreleptin was negative for genotoxicity in all the genotoxicity studies, and the applicant considered that metreleptin is not genotoxic.

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

Although no carcinogenicity studies of metreleptin had been performed, an immunohistochemical analysis of each tissue obtained from the 28-day subcutaneous toxicity study in mice (4.2.3.1-01) and 1-, 3-, and 6-month subcutaneous toxicity study in dogs (4.2.3.2-02) was conducted to evaluate carcinogenicity by detection of proliferating cell nuclear antigens (PCNAs). Metreleptin was considered to have low potential of carcinogenicity based on the following findings: (a) no cell proliferative activity had been observed, (b) the physiological action and effect had been almost similar in both metreleptin and endogenous leptin, and (c) no proliferative lesions had been reported in the long-term subcutaneous toxicity studies (4.2.3.2-01 and 4.2.3.2-02).

3.(iii).A.(3).1) Analysis of cell proliferative effect in mice (4.2.3.4-01 to -02)

In the 28-day subcutaneous toxicity study in mice (4.2.3.1-01), each tissue (the liver [liver cells and vascular endothelial cells[40]], kidney, lung, spleen, adrenal gland, pancreas, thyroid gland, stomach, duodenum, colon, testis [male mice], mammary gland [female mice], brain [including hypothalamus], and pituitary gland) was obtained from the animals in the control group and metreleptin groups at the end of the administration period. An immunohistochemical analysis was performed by avidin biotin-peroxidase complex method (ABC method) using anti-PCNA antibodies. There was no increase in the number of PCNA-positive cells or mitotic cells in the metreleptin groups, and the applicant considered that metreleptin does not have a cell proliferative effect in mice.

3.(iii).A.(3).2) Analysis of cell proliferative effect in dogs (4.2.3.4-03)

In the 1-, 3-, and 6-month subcutaneous toxicity study in dogs (4.2.3.2-02), each tissue (the liver [liver cells and vascular endothelial cells[40]], kidney, lung, spleen, adrenal gland, pancreas, thyroid gland, stomach, duodenum, colon, mammary gland [only female dogs], testis [only male dogs], brain [including hypothalamus], and pituitary gland) was obtained from the animals in the control group and 5 mg/kg/day group at the end of each administration period (n = 3/sex/group). An immunohistochemical analysis was

[40] Based on a report on leptin's proliferative effect on vascular endothelial cells (Bouloumie A, et al. *Circulation Research.* 1988;83:1059-66), the cell proliferative activity was assessed in hepatic vascular endothelial cells.

performed by ABC method using anti-PCNA antibodies. There was no increase in the number of PCNA-positive cells or mitotic cells in the metreleptin groups, and the applicant considered that metreleptin does not have a cell proliferative effect in dogs.

3.(iii).A.(4) Reproductive and developmental toxicity

A study of fertility and early embryonic development to implantation in mice, an embryo-fetal development study in mice, and a mouse study on pre- and postnatal development, including maternal function, were performed. Since an embryo-fetal toxicity study in rabbits (dose-finding study) had indicated no pharmacological action (such as inhibition of body weight gain), the applicant considered that it is difficult to evaluate embryo-fetal effect in rabbits. The reproductive and developmental toxicity were evaluated only in mice in which the pharmacological action had been confirmed since metreleptin and endogenous leptin were considered to have almost similar physiological action and effect.

3.(iii).A.(4).1) Study of fertility and early embryonic development to implantation in mice (4.2.3.5-01)

In this study, the vehicle[30] or metreleptin (1, 10, 30 mg/kg) was administered subcutaneously once daily[41] to male and female mice (CD-1, n = 25/sex/dose). No deaths occurred in both male and female mice and metreleptin treatment did not cause any effect on mating rate, conception rate, sperm test, number of days required for mating, reproductive organ weight, histopathological findings of testis and epididymis, and embryo viability. For male mice, dose-dependent inhibition of body weight gain was observed in the ≥1 mg/kg/day groups throughout the study period. Food consumption decreased at Week 1 in the ≥10 mg/kg/day groups and tended to decrease in the 30 mg/kg/day group until the end of the administration period. For female mice, inhibition of body weight gain and a decrease in food consumption at an early stage of administration were observed in the metreleptin groups. Based on the above, the NOAEL was determined to be 30 mg/kg/day for maternal and paternal toxicity and embryonic development under the conditions of this study.

3.(iii).A.(4).2) Embryo-fetal development study in mice (4.2.3.5-02)

In this study, the vehicle[30] or metreleptin (1, 10, 30 mg/kg) was administered subcutaneously once daily to mated female mice (CD-1, n = 25 per group) from gestation day 6 to 15. Although abortion was observed in 1 mouse in the 10 mg/kg/day group on gestation day 16, no abortions occurred in the 30 mg/kg/day group, indicating that the abortion observed was not due to metreleptin treatment. In the metreleptin groups, inhibition of body weight gain and a decrease in food consumption in the late stage of the administration period were observed in maternal animals during the administration period. Metreleptin treatment did not cause any effect on clinical signs, necropsy findings, and uterine weight of maternal animals and viability, intrauterine growth, sex ratio, external observations, internal organs, and skeletal structure of fetuses. Based on the above, the NOAEL was determined to be 30 mg/kg/day for maternal animals and embryos and fetuses under the conditions of this study.

[41] Male mice were administered metreleptin from 28 days before mating to the day before autopsy throughout the mating period, and female mice from 14 days before mating to gestation day 6 throughout the mating period.

3.(iii).A.(4).3) Dose-finding study on rabbit embryo-fetal toxicity study (Reference data 4.2.3.5-06)

In this study, the vehicle[34] or metreleptin (0.3, 1, 10, 30 mg/kg) was administered subcutaneously once daily to mated female rabbits (NZW, n = 5 per group) from gestation day 6 to 20. No deaths were reported and mild erythema and edema were observed at the injection site in the ≥10 mg/kg/day groups. Metreleptin treatment did not cause any effect on body weight, food consumption, and necropsy findings of maternal animals and viability, intrauterine growth, and sex ratio of embryos and fetuses. For external abnormalities, thoracoceloschisis, spina bifida, acrania, carpal flexion, short limb, adactyly, and brachydactyly were noted in 1 fetus in the 30 mg/kg/day group, and these were considered naturally occurring variations. Based on the above, the NOAEL was determined to be 30 mg/kg/day for maternal animals and embryos and fetuses under the conditions of this study.

3.(iii).A.(4).4) Mouse study on pre- and postnatal development, including maternal function (4.2.3.5-03)

In this study, the vehicle[34] or metreleptin (3, 10, 30 mg/kg) was administered subcutaneously once daily to mated female mice (CD-1, n=25 per group) from gestation day 6 to day 20 of nursing. In the metreleptin groups, deaths and moribund condition were observed (2 mice in the 3 mg/kg/day group, 1 mouse in the 10 mg/kg/day group, and 3 mice in the 30 mg/kg/day group). In the metreleptin groups, inhibition of body weight gain during the gestation period, a decreasing tendency in body weight during the nursing period, a decrease in food consumption during the administration period, dark red gastric and intestinal contents were observed for maternal animals, and abnormal parturition including deaths in the whole litter and intrauterine retention were observed; whole embryo resorptions were observed in the ≥10 mg/kg/day groups. A decrease in the number of alive pups, a trend toward low survival rates at 1 and 4 days post-birth, and inhibition of body weight gain at 1 and 21 days post-birth were observed in F1 pups in the metreleptin groups. For postweaning animals, deaths occurred in 1 male mouse (23 days post-birth) in the 10 mg/kg/day group, and 1 female mouse (25 days post-birth) in the 30 mg/kg/day group, and 1 male mouse (25 days post-birth) in the 30 mg/kg/day group was in moribund condition. In the metreleptin groups, a trend toward inhibition of body weight gain was observed and preputial separation (male mice) and delayed vaginal opening (female mice) were observed in the 30 mg/kg/day group. Metreleptin treatment did not cause any effect on fertility in F1 mice and viability, intrauterine growth, sex ratio, and external morphologies in F2 fetuses. Based on the above, the F0 and F1 maternal NOAEL for general and reproductive toxicity and the F1 pup NOAEL for developmental toxicity were determined to be <3 mg/kg/day, and the F1 mouse NOAEL for reproductive toxicity and the F2 fetal NOAEL for developmental toxicity were determined to be 30 mg/kg/day under the conditions of this study.

3.(iii).A.(4).5) Mouse study on pre- and postnatal development, including maternal function (additional study) (4.2.3.5-04)

An additional study was performed to examine the effect of metreleptin on maternal animals at the end of gestation and at the time of parturition as indicated in the mouse study on pre- and postnatal development,

including maternal function (4.2.3.5-03). In this study, the vehicle[42] or metreleptin 10 mg/kg was administered subcutaneously once daily to mated female mice (CD-1, n = 50 per group) for different treatment durations (gestation day 6-15, gestation day 15-18, or gestation day 6-18).[43] Deaths due to parturition or moribund condition were reported in the natural parturition group (the control group, 2 mice; the gestation day 15 to 18 group, 1 mouse; the gestation day 6 to 18 group, 4 mice). All fetal death occurred on day 1 of nursing in the gestation day 15 to 18 group (1 mouse) and on day 0 to 4 of nursing in the gestation day 6 to 18 group (9 mice); prolonged gestation period and increased incidence of abnormal parturition were also observed in the gestation day 6 to 18 group (9 mice). Inhibition of body weight gain during the administration period was observed in maternal animals in the metreleptin group, and body weight tended to decrease during the nursing period in the mice receiving metreleptin after gestation day 15. Decreased food consumption was observed from the administration period to the early stage of nursing in the gestation day 6 to 18 group and during the administration period in the gestation day 15 to 18 group. In the gestation day 15 to 18 group and the gestation day 6 to 18 group of the natural parturition group, the number of F1 pups of the same litter and alive pups and survival rates at 4 days post-birth tended to decrease or have a low number and there was a trend toward inhibition of body weight gain at 1 day post-birth. The prolonged gestation period, increased incidence of abnormal parturition, and effects on F1 pups were considered to be changes associated with exacerbation of nutritional status in maternal animals, which were likely to be prominent when metreleptin was administered during the period including the end of gestation (gestation day 15 to 18).

3.(iii).A.(5) Local tolerance (4.2.3.6-01)

In this study, metreleptin (20 mg/mL [1 mL/site], 5 mg/mL [4 mL/site]) was administered subcutaneously in a single dose to male and female rabbits (NZW; 20 mg/mL group, [n = 3/sex]; 5 mg/mL group, [4 males and 2 females]). The injection site was observed for up to 14 days after administration at the longest and histopathological studies were performed on Days 3 and 14 (n=3/day/dose). In the 20 mg/mL group, moderate hemorrhage at the injection site and chronic active inflammation were observed in 1 rabbit on Day 3, but no metreleptin-related changes were observed in other rabbits in the same group and 5 mg/mL group. The above results indicated that metreleptin 20 and 5 mg/mL had no apparent effect on the local tolerance on rabbit subcutaneous tissue.

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

3.(iii).A.(6).1) Antigenicity study (4.2.3.7-01)

The antigenicity of metreleptin 2.5 and 5 mg/mL was evaluated in an active systemic anaphylaxis test (ASA test) and a passive cutaneous anaphylaxis test (PCA test) using male guinea pigs (Dunkin-Hartley; n = 5 per group). In the ASA test, subcutaneous mass/necrosis associated with local inflammatory response at the subcutaneous injection site was observed in the group of guinea pigs receiving Freund's Complete Adjuvant (FCA). Severe anaphylaxis occurred in all bovine serum albumin (BSA)- and metreleptin 2.5 and 5

[42] 0.01% Tween 20, 10 mmol/L glutamic acid containing 2 % glycine

[43] In the control group, vehicle was administered on gestation day 6 to 18. In all groups, an embryo-fetal study and external observations were performed in half of the female mice after cesarean section on gestation day 18, and an autopsy was performed on the remaining female mice after they had nursed pups for 21 days after spontaneous parturition.

mg/mL-sensitized groups. In the PCA test, the most severe PCA occurred in guinea pigs treated with BSA and a dose-dependent reaction (stronger reaction was observed in guinea pigs treated with concomitant FCA at the same concentration) was observed in the metreleptin groups. The above results demonstrated that metreleptin induced acute systemic anaphylaxis and passive cutaneous anaphylaxis in guinea pigs. Anaphylaxis is considered very unlikely to be relevant to humans since it is a reaction commonly observed with animals immunized with heteroprotein.

3.(iii).A.(6).2) Blood compatibility study (4.2.3.7-02)

The hemolytic properties of metreleptin were evaluated using rat and human anticoagulated blood.[44] When rat and human blood was treated with metreleptin 125, 250, 500, and 1000 μg/mL, the hemolytic index did not exceed 5% at any concentration in human blood. On the other hand, metreleptin was considered to be hemolytic in rat blood since the hemolytic index was ≥5% at metreleptin 1000 μg/mL.

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

3.(iii).B.(1) Hemorrhage observed in several organs

PMDA asked the applicant to explain the mechanism of hemorrhage in several organs (gingiva, stomach, duodenum, bladder, etc.) observed in the 1-, 3-, and 6-month subcutaneous toxicity study in dogs (4.2.3.2-02) and potential safety concerns in humans.

The applicant responded as follows:

In the observation of clinical signs at the end of the 1-month treatment, hemorrhage was observed in the sclera (5 mg/kg/day group, 3 of 14 male dogs and 2 of 14 female dogs) and gingiva (1.5 mg/kg/day group, 2 of 14 female dogs; 5 mg/kg/day group, 4 of 14 male dogs and 2 of 14 female dogs). In the gross examination at necropsy, hemorrhage was observed in the gingiva (5 mg/kg/day group, 1 of 3 male dogs and 1 of 3 female dogs), bladder smooth muscle (5 mg/kg/day group, 1 of 3 male dogs), and smooth muscle of the gastrointestinal tract (duodenum, cecum, and colon) (5 mg/kg/day group, 1 of 3 male and/or female dogs). In the histopathological examination, hemorrhage was observed in the bladder submucosa or muscle layers (5 mg/kg/day group, 2 of 3 male dogs and 1 of 3 female dogs) and gastric submucosa (5 mg/kg/day group, 2 of 3 male dogs and 2 of 3 female dogs). In the observation of clinical signs at the end of the 3-month treatment, hemorrhage was observed in the gingiva (0.15 mg/kg/day group, 1 of 6 female dogs; 0.5 mg/kg/day group, 4 of 9 female dogs; 1.5 mg/kg/day group, 4 of 9 male dogs and 5 of 9 female dogs; 5 mg/kg/day group, 5 of 9 male dogs and 6 of 9 female dogs). Hemorrhage in the gastrointestinal mucosa and submucosa was considered to be due to the effect of stress related to a decrease in food consumption. Since no decrease in platelet count or prolongation of clotting time was observed, there is no possibility that hemorrhage may have been caused by the effects of metreleptin on the blood coagulation system. On the other hand, leptin is known to have angiogenic activity.[45] Since it is inferred that vascularized terminal capillary vessels create an environment in which hemorrhage tends to occur, the angiogenic activity of metreleptin may have induced

[44] Metreleptin was allowed to react with human or rat anticoagulated blood and left for 4 hours at 37°C. After absorbance was measured at 545 nm, the quantity of partial hemolysis, an indicator of hemoglobin levels, was determined using 0% of hemolysis rate and complete hemolysis as comparators.

[45] Bouloumie A, et al. *Circulation Research.* 1988;83:1059-66. Cao R, et al. *Proc Natl Acad Sci USA.* 2001;98:6390-5

hemorrhage in the gingiva, etc. Therefore, the possibility that similar hemorrhage episodes may occur in humans cannot be ruled out completely. However, all hemorrhagic findings from dogs, except local gingival hemorrhage in the 0.15 to 1.5 mg/kg/day groups, are reported only in the 5 mg/kg/day group, and the local gingival hemorrhage observed in the 0.15 to 1.5 mg/kg/day groups is found at the time of clinical observations, suggesting that it is a transient change. Hence, this finding is considered to be low toxicological significance. Furthermore, the analysis of hemorrhagic adverse events in the Japanese and foreign studies demonstrated that the number of subjects with hemorrhagic adverse events was low in the Japanese investigator-initiated trial (Study KUTR-003-1, 2 of 4 subjects), Japanese clinical research (Study KUTR-003-0, 1 of 11 subjects), Japanese investigational study[46] (Study KUTR-003-2, 1 of 12 subjects), foreign NIH clinical study (Study [redacted]1265/20[redacted]0769, 2 of 55 subjects), and foreign Amylin-sponsored study (Study FHA101, 1 of 10 subjects), INTEGRATED SUMMARY OF SAFETY (12 of 785 subjects)[47], and most of the adverse events were mild in severity and was reversible. The above results indicate that local hemorrhage related to the angiogenic activity of metreleptin is unlikely to be clinically significant.

PMDA considers as follows:
Hemorrhage is unlikely to cause major problems in clinical use because (a) all hemorrhagic findings from the study in dogs, except gingival hemorrhage, are reported only in the 5 mg/kg/day group after 1-month treatment, and the level of exposure ($AUC_{0\text{-}24h}$)[48] in this group at this timepoint (32,981 ng·h/mL for male dogs and 64,817 ng·h/mL for female dogs) is estimated to be approximately 10 times the level of exposure ($AUC_{0\text{-}24h}$)[49] in the clinical studies, (b) no clinical data are available for studies with a control group, and (c) the association of hemorrhagic adverse events with metreleptin treatment has not been clarified in humans. On the other hand, given that gingival hemorrhage occurred below the level of exposure ($AUC_{0\text{-}24h}$)[49] in the clinical studies and the tissue distribution of metreleptin has not been demonstrated in animals and humans and that the association of this event with the pharmacological action of metreleptin has not been denied, it is necessary to provide the information on hemorrhage induced by metreleptin treatment in the toxicity studies in the package insert.

3.(iii).B.(2) Effect on the gastrointestinal tract

PMDA asked the applicant to explain the mechanism of gastrointestinal lesions in mice and dogs (gastric mucosal erosion in mice, ulcers on the gastric mucosa and intestinal mucosal hemorrhage in dogs, etc.).

The applicant responded as follows:
The incidence of gastric mucosal erosion observed in the 3- and 6-month subcutaneous toxicity study in mice (4.2.3.2-01) tended to be slightly higher in female mice than in male mice, and a decrease in food

46 This is a clinical research approved for implementation that was reviewed and reported in the Investigational Medication Evaluation Committee in May 2010 and the Advanced Medicine Expert Committee and Central Social Insurance Medical Council in July 2010.
47 The major hemorrhagic transformation included epistaxis, bloody stool, hematuria, vaginal hemorrhage, and ecchymosis. Hemorrhagic transformation related to the injection site (such as injection site hemorrhage) is excluded.
48 The extent of exposure on Day 7 for male dogs and Day 14 for female dogs was provided because of the effect of antibody production on the measurement system.
49 Compared with the maximum $AUC_{0\text{-}\tau}$ (τ, 24 h) (3385 ng·h/mL) in the Japanese physician-driven trial (Study KUTR-003-001) of Japanese patients with lipodystrophy.

consumption (a maximum decrease of 15% in the 30 mg/kg/day group) in the metreleptin groups was observed at the end of the 3-month treatment. In the 1-, 3-, and 6-month subcutaneous toxicity study in dogs (4.2.3.2-02), an examination at the end of 1-month treatment showed gastric mucosal hemorrhage in 2 of 3 male dogs and 1 of 3 female dogs and gastric and/or intestinal purulent inflammation in 2 of 3 male dogs and 2 of 3 female dogs in the 5 mg/kg/day group and a decrease in food consumption (a maximum decrease of 63% in the 5 mg/kg/day group) in the ≥0.5 mg/kg/day groups. In a feeding suppression study using SD rats (SD rats were fed with a restricted diet designed to have 75%, 50%, and 25% of the calories in the fully-fed group and they were reared for 2 weeks), a reddish change or blood-stained contents in the glandular stomach or erosion of the glandular stomach were observed. The erosion of the glandular stomach was likely to be induced by the release of corticosteroid caused by stress due to feeding restriction (Levin S, et al. *Toxicologic Pathology.* 1993;21:1-14). In general, adrenocortical hormone is considered to increase gastric acid secretion and reduce mucosal regeneration (*Toxicological Histopathology.* Japanese Society for Toxicologic Pathology 2000;156-7). A decrease in food consumption was observed in male mice until around 2 months after administration, but food consumption continued to decrease in female mice even thereafter. The blood chemistry test revealed a marked decrease in glucose, cholesterol, triglyceride, etc., strongly suggesting the effect of decreased food consumption. In the group of dogs with gastrointestinal effect, a decrease in food consumption and changes in blood chemistry parameters were observed, as with the mouse study. The above results indicate that the gastric mucosal erosion in mice and gastrointestinal mucosal hemorrhage, ulcers, and purulent inflammation in dogs are attributed to stress-related mucosal injury caused by a marked decrease in food consumption and changes secondary to it.

PMDA considers as follows:
As for the effect on the gastrointestinal mucosa in mice and dogs, besides the mechanism explained by the applicant, a decrease in food consumption in the 10 mg/kg/day group was not more than 13% in mice at the end of the 3-month treatment. In contrast, gastric mucosal erosion was observed in all female mice in the 10 mg/kg/day group and the incidence was higher compared with male mice. Changes in various blood chemistry parameters related to lipid metabolism which is the pharmacological action of metreleptin, were more significant in female mice, and exacerbation of conditions associated with these changes might have affected gastrointestinal mucosal injury. On the other hand, decreased food consumption was highly associated with gastrointestinal mucosal injury and these findings were found to be reversible; therefore, they were considered to be unlikely to be clinically significant.

3.(iii).B.(3) Perivasculitis

Because perivasculitis had been observed in all examinations at the end of the 1-, 3-, and 6-month treatment periods in the 1-, 3-, and 6-month subcutaneous toxicity study in dogs (4.2.3.2-02), PMDA asked the applicant to explain the toxicological significance of this finding and potential safety concerns in humans.

The applicant responded as follows:
Because perivasculitis observed at all examination timepoints occurred only at the injection site and it was sporadically observed also in the control group, perivasculitis at the injection site is considered to be changes

related to injection techniques. On the other hand, of dogs in which perivasculitis was observed at the sites other than the injection site at the end of the 1-month treatment (0.05 mg/kg/day group, 1 of 3 female dogs; 0.15 mg/kg/day group, 1 of 3 male dogs; 0.5 mg/kg/day group, 1 of 3 female dogs; 5 mg/kg/day group, 2 of 3 male dogs and 2 of 3 female dogs), 2 of 3 male dogs and 1 of 3 female dogs in the 5 mg/kg/day group had perivasculitis in several organs. Idiopathic polyangiitis (perivasculitis) is known to develop spontaneously in dogs.[50] While drug-induced vasculitis (perivasculitis) is often localized to the coronary artery and followed by hemorrhage of the vascular wall, spontaneous vasculitis (perivasculitis) is not localized to the coronary artery but to smaller arteries and not associated with hemorrhage (Clemo FAS, et al. *Toxicologic Pathology.* 2003;31:25-31). In this study, perivasculitis was rarely observed in the coronary artery and there were no findings associated with hemorrhage, indicating correspondence with spontaneous vasculitis. On the other hand, when compared with background data from the laboratory over a certain period of time (19[redacted] to 20[redacted]), including the study period for this study (19[redacted] to 19[redacted]), perivasculitis occurred more frequently and in various organs in this study and therefore the possibility that metreleptin treatment might have had an impact on the increase in the incidence of spontaneous lesions cannot be ruled out. However, perivasculitis was not reported at 3 months post-dose and after recovery in this study, in the 28-day subcutaneous toxicity study in dogs (4.2.3.2-06), and in the 4-week intravenous toxicity study in dogs (4.2.3.2-08), and perivasculitis-related adverse events have not occurred in the Japanese studies, suggesting that it is unlikely to be a major problem in clinical practice.

PMDA considers as follows:

Although perivasculitis observed in this study is characterized by spontaneous lesions, there are evident increases in the incidence and the number of sites of perivasculitis in the 5 mg/kg/day group. In this study, perivasculitis tended to occur commonly in adipose tissue but the literatures presented have not reported an occurrence of perivasculitis in adipose tissue. From these findings, the association of the event with metreleptin treatment cannot be denied. Because inflammatory cells observed in this study were mainly composed of lymphocytes and plasma cells and leptin has been reported to affect the immune system (Lord GM, et al. *Nature.*1998;394:897-901), the leptin's effect on the immune system may have contributed to the development of perivasculitis. On the other hand, with respect to the characteristics of inflammatory cells in spontaneous vasculitis (perivasculitis), the literatures presented have reported that the inflammatory cells are mainly composed of monocytes and other components such as lymphocytes, plasma cells, and macrophages (Ruben Z, et al. *Toxicologic Pathology.* 1989;17:145-52), and there is also a literature suggesting the association of spontaneous vasculitis in dogs with immunity (Snyder P, et al. *Veterinary Pathology.* 1995:32:337-345); therefore, metreleptin's effect on the immune system could have caused significant perivasculitis in dogs predisposed to vasculitis (perivasculitis) mediated by the immune system. Based on the fact that perivasculitis was not reported in mice and there was no increase in inflammation-related markers in the Japanese studies, these findings are of little relevance to humans and perivasculitis was considered unlikely to be a problem in clinical practice.

[50] Ruben Z, et al. *Toxicologic Pathology.* 1989;17:145-52. Maxie MG, et al. *Pathology of Domestic Animals. 5th ed*. ed. by Maxie MG et al. Elsevier, Philadelphia, 2007;69-72

4. Clinical data

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

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

Enzyme immunoassay and radioimmunoassay were used to determine the concentrations of human serum leptin and the lower limits of quantification were 0.040 and 0.5 ng/mL, respectively. Biosensor immunoassay was used to measure the serum levels of antibodies against metreleptin and the levels of serum neutralizing antibodies against metreleptin were determined by bioassay. Since the antibodies used for enzyme immunoassay and radioimmunoassay can not distinguish between endogenous leptin versus metreleptin, measured values are expressed as the total of both levels.

4.(ii) Summary of clinical pharmacology studies

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

As the evaluation data, the results from a Japanese investigator-initiated trial in Japanese patients with lipodystrophy (Study KUTR-003-1) were submitted. As the reference data, the results from a foreign phase I intravenous study in foreign healthy adult subjects (Study LEPT-[redacted]0121), a foreign phase I subcutaneous study (Study LEPT-[redacted]0272), a Japanese clinical research in Japanese patients with lipodystrophy (Study KUTR-003-0), and a foreign NIH clinical study in foreign patients with lipodystrophy (Study [redacted]1265/20[redacted]0769) were submitted. The results from the main studies are described below. The drug products used in each study were as shown in Table 8 and a lyophilized formulation refers to a "drug product" in the following sections.

Table 8. Drug products used in each study

Name of the study or research (study number)	Drug product	Metreleptin concentration (at the time of preparation)
Japanese investigator-initiated trial (Study KUTR-003-1)	Lyophilized formulation	5 mg/mL
Japanese clinical research (Study KUTR-003-0)		
Japanese investigational study (Study KUTR-003-2)		
Foreign NIH clinical study (Study [redacted]1265/20[redacted]0769)		
Foreign Amylin-sponsored study (Study FHA101)		
Foreign phase I intravenous study (Study LEPT-[redacted]0121)	Frozen formulation[a)]	5 mg/mL and 20 mg/mL
Foreign phase I subcutaneous study (Study LEPT-[redacted]0272)		

a) A solution of metreleptin for injection was cryopreserved and it was thawed before use.

4.(ii).A.(1) Studies in healthy adult subjects

4.(ii).A.(1).1) Phase I intravenous study (Reference data 5.3.3.1-1, Study LEPT-[redacted]0121 [[redacted] to [redacted] 19[redacted]])

A randomized, double-blind, placebo-controlled, parallel-group, comparative study was conducted in foreign healthy adult subjects (target sample size from 96 to 264[51]) to evaluate the safety and pharmacokinetics of a frozen formulation of metreleptin following multiple intravenous administration.

[51] To minimize the potential for assigning subjects to receive an unsafe dose or a safe but ineffective dose in this study, the number of subjects assigned to each dose group could depend on results; therefore, the planned minimum and maximum number of subjects was provided.

In the fixed-dose group, placebo, a dose of 0.3, 1.0, and 3.0 mg/kg of metreleptin (5 mg/mL formulation), or a dose of 0.3 mg/kg of metreleptin (20 mg/mL formulation) was to be administered intravenously once daily for 28 days. In the dose-titration group, metreleptin (20 mg/mL formulation) was to be administered intravenously once daily at a dose of 0.1 mg/kg for 10 days, followed by a dose of 0.3 mg/kg for 10 day, then by a dose of 0.6 mg/kg for 10 days (30 days in total), or at a dose of 0.3 mg/kg for 10 days, followed by a dose of 0.6 mg/kg for 10 days, then by a dose of 1.0 mg/kg for 10 days (30 days in total). Forty-two subjects were assigned to the placebo group, 67 subjects to the fixed-dose group (16 subjects in the 0.3 mg/kg group of 5 mg/mL formulation, 16 subjects in the 0.3 mg/kg group of 20 mg/mL formulation, 18 subjects in the 1.0 mg/kg group of 5 mg/mL formulation, and 17 subjects in the 3.0 mg/kg group of 5 mg/mL formulation), and 16 subjects to the dose-titration group (6 subjects in the 0.1-0.6 mg/kg group of 20 mg/mL formulation and 10 subjects in the 0.3-1.0 mg/kg group of 20 mg/mL formulation).[52]

All of 124 subjects receiving the study drug (42 subjects in the placebo group and 82 subjects in the metreleptin group) were included in the safety analysis set. The pharmacokinetic analysis set included 109 subjects (the fixed-dose group, 33 subjects in the placebo group and 53 subjects in the metreleptin group; the dose-titration group, 8 subjects in the placebo group and 15 subjects in the metreleptin group) on Day 1, and 87 subjects (33 subjects in the placebo group and 54 subjects in the metreleptin group) on Day 15 (only the fixed-dose group). There were 14 subjects who were withdrawn, from the trial and they included 1 subjects in the placebo group (other reasons) and 13 subjects in the metreleptin group (adverse events [investigator's decision] for 6 subjects, adverse events [subject's decision] for 3 subjects, other reasons for 3 subjects, and lost to follow-up for 1 subject).

For pharmacokinetics, pharmacokinetic parameters in the fixed-dose group on Day 1 were as shown in Table 9.

Table 9. Pharmacokinetic parameters in the fixed-dose group on Day 1

Treatment group (N)[a)]	C_{max} (ng/mL)	$AUC_{0\text{-}inf}$ (ng·h/mL)	$t_{1/2,z}$ (h)	CL (mL/h/kg)
Placebo group (n = 33)	13.9 ± 13.6	272.2 ± 271.2[b)]	-	-
0.3 mg/kg group (n = 27)	6160 ± 1386	3909 ± 754	3.34 ± 1.88	79.6 ± 16.1
1.0 mg/kg group (n = 14)	17,582 ± 2914	12,545 ± 2449	3.41 ± 0.85	82.5 ± 15.6
3.0 mg/kg group (n = 12)	32,887 ± 4976	32,776 ± 7251	3.42 ± 0.82	95.8 ± 21.6

Mean ± SD; -, not calculated;
C_{max}, maximum serum concentration; $AUC_{0\text{-}inf}$, area under the serum concentration-time curve extrapolated to infinity; $t_{1/2,z}$, terminal elimination half-life; CL, clearance
a) N represents the number of subjects who used a 5 mg/mL formulation; however, the number of subjects in the 0.3mg/kg group denotes the sum of those using 5 mg/mL and 20 mg/mL formulations.
b) $AUC_{0\text{-}24h}$

The mean ratios of C_{max} and AUC ($AUC_{0\text{-}inf}$ for Day 1 and $AUC_{0\text{-}\tau}$ for Day 15) on Day 15 to Day 1 in the fixed-dose group (Day 15/Day 1) were calculated. In the 0.3, 1.0, and 3.0 mg/kg group, the values were 0.9, 0.9, and 1.0 for C_{max}, respectively, and 1.9, 1.0, and 1.0 for AUC, respectively.

For safety, the occurrence of adverse events and adverse drug reactions was as shown in Table 10.

[52] After being assigned to any of the 4 groups ("20.0-23.4 kg/m^2," "23.5-27.5 kg/m^2," "27.6-30.0 kg/m^2," or "30.1-36.0 kg/m^2") according to BMI, subjects were assigned to each treatment group.

Table 10. Occurrence of adverse events and adverse drug reactions[a)b)]

		Placebo (n = 42)	5 mg/mL formulation (mg/kg)			20 mg/mL formulation (mg/kg)		
			0.3 (n = 16)	1.0 (n = 17)	3.0 (n = 17)	0.1-0.6 (n = 6)	0.3 (n = 16)	0.3-1.0 (n = 10)
Any adverse event		31 (74)	14 (88)	15 (88)	15 (88)	5 (83)	16 (100)	10 (100)
Any adverse drug reaction		21 (50)	13 (81)	12 (71)	14 (82)	5 (83)	15 (94)	9 (90)
Headache	Adverse event	14 (33)	5 (31)	7 (41)	10 (59)	4 (67)	9 (56)	10 (100)
	Adverse drug reaction	13 (31)	5 (31)	3 (18)	7 (41)	4 (67)	8 (50)	9 (90)
Pyrexia	Adverse event	2 (5)	0 (0)	1 (6)	1 (6)	3 (50)	9 (56)	9 (90)
	Adverse drug reaction	2 (5)	0 (0)	1 (6)	0 (0)	3 (50)	7 (44)	8 (80)
Anorexia	Adverse event	5 (12)	5 (31)	3 (18)	5 (29)	0 (0)	4 (25)	3 (30)
	Adverse drug reaction	5 (12)	5 (31)	3 (18)	5 (29)	0 (0)	4 (25)	3 (30)
Contact erythema	Adverse event	4 (10)	2 (13)	2 (12)	2 (12)	2 (33)	9 (56)	1 (10)
	Adverse drug reaction	2 (5)	2 (13)	2 (12)	2 (12)	2 (33)	8 (50)	0 (0)
Chills	Adverse event	3 (7)	0 (0)	0 (0)	1 (6)	1 (17)	8 (50)	8 (80)
	Adverse drug reaction	3 (7)	0 (0)	0 (0)	0 (0)	1 (17)	8 (50)	7 (70)

N (%)

a) Adverse events with an incidence of ≥20% in the metreleptin groups
b) A significant digit for an incidence is provided as described in the clinical study report.

No deaths or serious adverse events were reported. Adverse events leading to discontinuation occurred in 2 subjects in the 0.3 mg/kg group of 5 mg/mL formulation (rash/pruritus, rash erythematous; 1 subject each), 1 subject in the 1.0 mg/kg group of 5 mg/mL formulation (urticaria/pruritus), 4 subjects in the 3.0 mg/kg group of 5 mg/mL formulation (rash [2], pruritus[1], oedema peripheral [1]), 1 subject in the 0.3 mg/kg group of 20 mg/mL formulation (pyrexia/asthenia/chills), 1 subject in the 0.3 to 1.0 mg/kg groups of 20 mg/mL formulation (pyrexia/chills). There were no clinically significant changes in laboratory tests, vital signs, or ECG.

When anti-leptin antibodies were determined on Days 1, 11, 15, 29, and 31, the percentages of subjects who were tested positive for anti-leptin antibodies at any of the timepoints were 0% in the placebo group, 23% (11 of 47 subjects [2 subjects in the 0.3 mg/kg group, 3 subjects in the 1.0 mg/kg group, and 6 subjects in the 3.0 mg/kg group]) in the 5 mg/mL formulation group, and 84 % (27 of 32 subjects [4 subjects in the 0.1-0.6 mg/kg group, 15 subjects in the 0.3 mg/kg group, and 8 subjects in the 0.3-1.0 mg/kg group]) for the 20 mg/mL formulation group.

4.(ii).A.(1).2) Phase I subcutaneous study (Reference data 5.3.3.1-2, Study LEPT-[redacted]0272 [[redacted] 19[redacted] to [redacted] 19[redacted]])

A randomized, double-blind, placebo-controlled, parallel-group, comparative study was conducted in foreign healthy adult subjects (target sample size from 96 to 324[51]) to evaluate the safety and pharmacokinetics of a frozen formulation of metreleptin following multiple subcutaneous administration.

In the subcutaneous administration group, placebo, a dose of 0.01, 0.03, 0.1, and 0.3 mg/kg of metreleptin (5 mg/mL formulation), or a dose of 0.3 mg/kg of metreleptin (20 mg/mL formulation) was to be administered subcutaneously once daily for 4 weeks, and subjects with BMI of 27.6 to 36.0 kg/m^2 [53] were to continue to

[53] A subject with BMI of 20.0 to 27.5 kg/m^2 is defined as a "nonobese subject" and a subject with BMI of 27.6 to 36.0 kg/m^2 is defined as an "obese

receive the study drug subcutaneously for 20 weeks (24 weeks in total). In the continuous subcutaneous administration group, placebo or a dose of 0.3, 1.0, 2.0 mg/kg of metreleptin (5 mg/ml formulation) was to be administered continuously once daily over 20 ± 4 hours for 4 weeks, and subjects with BMI of 27.6 to 36.0 kg/m^2 were to continue to receive the study drug subcutaneously for 20 weeks (24 weeks in total). In the subcutaneous administration group, 44 subjects were assigned to the placebo group, 16 subjects to the 0.01 mg/kg group of 5 mg/mL formulation, 16 subjects to the 0.03 mg/kg group of 5 mg/mL formulation, 31 subjects to the 0.1 mg/kg group of 5 mg/mL formulation, 27 subjects to the 0.3 mg/kg group of 5 mg/mL formulation, and 7 subjects to the 0.3 mg/kg group of 20 mg/mL formulation. In the continuous subcutaneous administration group, 31 subjects were assigned to the placebo group, 27 subjects to the 0.3 mg/kg group of 5 mg/mL formulation, 29 subjects to the 1.0 mg/kg group of 5 mg/mL formulation, and 28 subjects to the 2.0 mg/kg group of 5 mg/mL formulation.[54]

All of 250 subjects receiving the study drug (subcutaneous administration group, 42 subjects in the placebo group and 96 subjects in the metreleptin group; continuous subcutaneous administration group, 31 subjects in the placebo group and 81 subjects in the metreleptin group) were included in the safety analysis set and pharmacokinetic analysis set. There were 61 subjects who were withdrawn, and they included 11 subjects in the placebo group (subject's request for 7 subjects, lost to follow-up for 2 subjects, protocol deviation for 1 subject, protocol violation for 1 subject) and 13 subjects in the metreleptin group (subject's request for 4 subjects, inability to receive treatment for 4 subjects, adverse events for 3 subjects, protocol violation for 1 subject, lost to follow-up for 1 subject) for the subcutaneous administration group as well as 4 subjects in the placebo group (subject's request for 3 subjects, protocol deviation for 1 subject) and 33 subjects in the metreleptin group (subject's request for 17 subjects, adverse events for 12 subjects, protocol violation for 3 subjects, protocol deviation for 1 subject) for the continuous subcutaneous administration group.

The pharmacokinetic parameters in the subcutaneous administration group on Day 1 were as shown in Table 11.

Table 11. Pharmacokinetic parameters in the subcutaneous administration group on Day 1

Dose (mg/kg/day)		C_{max} (ng/mL)	t_{max} (h)	$AUC_{0\text{-inf}}$ (ng·h/mL)	$t_{1/2,z}$ (h)	CL/F (mL/h/kg)	Vz/F (mL/kg)
Placebo (n = 73)		21 ± 22	15.0 ± 5.7	850 ± 1166	-	-	-
5 mg/mL formulation	0.01 (n = 16)	14 ± 10	4.8 ± 3.9	131 ± 104	4.7 ± 3.0	137 ± 112	804 ± 634
	0.03 (n = 16)	37 ± 17	3.8 ± 1.2	337 ± 132	4.7 ± 2.0	106 ± 53	715 ± 447
	0.1 (n = 31)	119 ± 32	4.3 ± 1.5	1180 ± 267	4.3 ± 2.7	89 ± 20	519 ± 276
	0.3 (n = 26)	343 ± 104	4.0 ± 1.5	3657 ± 781	3.8 ± 1.4	86 ± 18	444 ± 117
20 mg/mL formulation	0.3 (n = 7)	207 ± 42	5.4 ± 2.4	2059 ± 504	3.1 ± 0.7	153 ± 34	674 ± 150

Mean ± SD; -, not calculated;
C_{max}, maximum serum concentration; t_{max}, time to reach maximum serum concentration; $AUC_{0\text{-inf}}$, area under the serum concentration-time curve extrapolated to infinity; $t_{1/2,z}$, terminal elimination half-life; CL/F, apparent clearance; Vz/F, apparent volume of distribution during the terminal phase

subject."

[54] The number of subjects with BMI of 20.0 to 27.5 kg/m^2 was 20, 8, 8, 13, 9, 4, 12, 9, 8, and 9, in the placebo (subcutaneous administration), 0.01, 0.03, 0.1, and 0.3 mg/kg (5 mg/mL formulation, subcutaneous administration), 0.3 mg/kg, (20 mg/mL formulation, subcutaneous administration), placebo (continuous subcutaneous administration), and 0.3, 1.0, and 2.0 mg/kg (5 mg/mL formulation, continuous subcutaneous administration) group, respectively, while the number of those with BMI of 27.6 to 36.0 kg/m^2 was 24, 8, 8, 18, 18, 3, 19, 18, 21, and 19, respectively.

For safety, the occurrence of adverse events and adverse drug reactions up to Week 4 in nonobese and obese subjects was as shown in Tables 12 and 13.

Table 12. Occurrence of adverse events and adverse drug reactions up to Week 4 in nonobese subjects[a)b)]

		Subcutaneous administration (mg/kg/day)						Continuous subcutaneous administration (mg/kg/day)			
		Placebo (n = 19)	5 mg/mL formulation				20 mg/mL formulation	Placebo (n = 12)	5 mg/mL formulation		
			0.01 (n = 8)	0.03 (n = 8)	0.1 (n = 13)	0.3 (n = 8)	0.3 (n = 4)		0.3 (n = 9)	1.0 (n = 8)	2.0 (n = 9)
Any adverse event		17 (89)	8 (100)	8 (100)	12 (92)	8 (100)	2 (50)	11 (92)	9 (100)	8 (100)	9 (100)
Any adverse drug reaction		8 (42)	4 (50)	7 (88)	11 (85)	7 (88)	2 (50)	9 (75)	8 (89)	8 (100)	9 (100)
Injection site ecchymosis	Adverse event	10 (53)	2 (25)	6 (75)	9 (69)	8 (100)	1 (25)	7 (58)	3 (33)	3 (38)	2 (22)
	Adverse drug reaction	4 (21)	1 (13)	4 (50)	9 (69)	7 (88)	1 (25)	3 (25)	1 (11)	0 (0)	1 (11)
Injection site erythema	Adverse event	4 (21)	0 (0)	5 (63)	8 (62)	4 (50)	2 (50)	9 (75)	8 (89)	8 (100)	8 (89)
	Adverse drug reaction	4 (21)	0 (0)	4 (50)	8 (62)	4 (50)	2 (50)	9 (75)	8 (89)	8 (100)	8 (89)
Headache	Adverse event	9 (47)	3 (38)	3 (38)	3 (23)	5 (63)	1 (25)	4 (33)	6 (67)	3 (38)	5 (56)
	Adverse drug reaction	1 (5)	2 (25)	2 (25)	2 (15)	1 (13)	1 (25)	1 (8)	4 (44)	2 (25)	3 (33)
Nausea	Adverse event	1 (5)	0 (0)	2 (25)	1 (8)	4 (50)	1 (25)	2 (17)	2 (22)	1 (13)	1 (11)
	Adverse drug reaction	1 (5)	0 (0)	1 (13)	1 (8)	0 (0)	1 (25)	1 (8)	2 (22)	1 (13)	0 (0)
Rhinitis	Adverse event	3 (16)	2 (25)	0 (0)	1 (8)	4 (50)	1 (25)	0 (0)	1 (11)	2 (25)	1 (11)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (13)	0 (0)
Injection site inflammation	Adverse event	0 (0)	0 (0)	0 (0)	0 (0)	2 (25)	2 (50)	4 (33)	8 (89)	8 (100)	7 (78)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	0 (0)	2 (25)	2 (50)	4 (33)	8 (89)	8 (100)	7 (78)
Injection site pruritus	Adverse event	1 (5)	0 (0)	0 (0)	3 (23)	1 (13)	1 (25)	3 (25)	7 (78)	8 (100)	7 (78)
	Adverse drug reaction	1 (5)	0 (0)	0 (0)	3 (23)	1 (13)	1 (25)	2 (17)	7 (78)	8 (100)	7 (78)
Dermatitis contact	Adverse event	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	8 (67)	6 (67)	5 (63)	6 (67)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	4 (33)	1 (11)	3 (38)	4 (44)
Injection site pain	Adverse event	4 (21)	1 (13)	2 (25)	2 (15)	1 (13)	1 (25)	5 (42)	3 (33)	6 (75)	7 (78)
	Adverse drug reaction	1 (5)	0 (0)	1 (13)	2 (15)	1 (13)	1 (25)	5 (42)	3 (33)	6 (75)	6 (67)
Injection site reaction	Adverse event	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	2 (17)	2 (22)	5 (63)	5 (56)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	2 (17)	1 (11)	5 (63)	3 (33)
Fatigue	Adverse event	1 (5)	2 (25)	1 (13)	0 (0)	0 (0)	0 (0)	1 (8)	3 (33)	3 (38)	3 (33)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (8)	1 (11)	2 (25)	1 (11)
Injection site mass	Adverse event	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (8)	2 (22)	2 (25)	4 (44)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (8)	2 (22)	2 (25)	4 (44)

N (%)

a) Adverse events with an incidence of ≥20% in the metreleptin groups for subcutaneous administration or continuous subcutaneous administration.
b) A significant digit for an incidence is as described in the clinical study report.

Table 13. Occurrence of adverse events and adverse drug reactions up to Week 4 in obese subjects[a)b)]

		Subcutaneous administration (mg/kg/day)						Continuous subcutaneous administration (mg/kg/day)			
		Placebo (n = 23)	5 mg/mL formulation				20 mg/mL formulation	Placebo (n =19)	5 mg/mL formulation		
			0.01 (n = 8)	0.03 (n = 8)	0.1 (n = 18)	0.3 (n = 18)	0.3 (n =3)		0.3 (n = 18)	1.0 (n = 19)	2.0 (n = 18)
Any adverse event		19 (83)	8 (100)	8 (100)	18 (100)	18 (100)	3 (100)	19 (100)	18 (100)	19 (100)	18 (100)
Any adverse drug reaction		10 (43)	6 (75)	3 (38)	15 (83)	18 (100)	3 (100)	13 (68)	18 (100)	19 (100)	18 (100)
Injection site ecchymosis	Adverse event	14 (61)	5 (63)	3 (38)	14 (78)	15 (83)	2 (67)	15 (79)	9 (50)	10 (53)	8 (44)
	Adverse drug reaction	5 (22)	1 (13)	2 (25)	8 (44)	7 (39)	1 (33)	3 (16)	3 (17)	0 (0)	4 (22)
Injection site erythema	Adverse event	2 (9)	0 (0)	1 (13)	13 (72)	16 (89)	3 (100)	8 (42)	18 (100)	18 (95)	17 (94)
	Adverse drug reaction	1 (4)	0 (0)	1 (13)	13 (72)	16 (89)	3 (100)	7 (37)	18 (100)	18 (95)	17 (94)
Headache	Adverse event	7 (30)	6 (75)	2 (25)	9 (50)	10 (56)	0 (0)	12 (63)	8 (44)	11 (58)	14 (78)
	Adverse drug reaction	1 (4)	2 (25)	1 (13)	2 (11)	7 (39)	0 (0)	6 (32)	4 (22)	5 (26)	11 (61)
Injection site inflammation	Adverse event	0 (0)	0 (0)	0 (0)	6 (33)	7 (39)	3 (100)	5 (26)	16 (89)	16 (84)	17 (94)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	6 (33)	7 (39)	3 (100)	5 (26)	16 (89)	16 (84)	17 (94)
Injection site pruritus	Adverse event	1 (4)	0 (0)	0 (0)	7 (39)	9 (50)	3 (100)	1 (5)	15 (83)	15 (79)	17 (94)
	Adverse drug reaction	1 (4)	0 (0)	0 (0)	7 (39)	9 (50)	3 (100)	1 (5)	15 (83)	15 (79)	17 (94)
Dermatitis contact	Adverse event	0 (0)	0 (0)	0 (0)	1 (6)	0 (0)	2 (67)	14 (74)	14 (78)	14 (74)	12 (67)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (33)	3 (16)	7 (39)	3 (16)	9 (50)
Injection site pain	Adverse event	3 (13)	0 (0)	0 (0)	2 (11)	5 (28)	1 (33)	6 (32)	6 (33)	8 (42)	15 (83)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	2 (11)	4 (22)	1 (33)	5 (26)	6 (33)	7 (37)	15 (83)
Injection site reaction	Adverse event	0 (0)	0 (0)	0 (0)	3 (17)	6 (33)	1 (33)	3 (16)	5 (28)	12 (63)	12 (67)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	3 (17)	6 (33)	1 (33)	0 (0)	5 (28)	12 (63)	11 (61)
Fatigue	Adverse event	3 (13)	1 (13)	0 (0)	2 (11)	4 (22)	0 (0)	2 (11)	1 (6)	5 (26)	5 (28)
	Adverse drug reaction	0 (0)	1 (13)	0 (0)	1 (6)	1 (6)	0 (0)	2 (11)	0 (0)	2 (11)	4 (22)
Injection site mass	Adverse event	0 (0)	0 (0)	0 (0)	0 (0)	1 (6)	1 (33)	1 (5)	4 (22)	3 (16)	9 (50)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	0 (0)	0 (0)	1 (33)	1 (5)	4 (22)	3 (16)	9 (50)
Injection site oedema	Adverse event	0 (0)	0 (0)	0 (0)	5 (28)	2 (11)	1 (33)	3 (16)	2 (11)	7 (37)	5 (28)
	Adverse drug reaction	0 (0)	0 (0)	0 (0)	4 (22)	2 (11)	1 (33)	2 (11)	2 (11)	7 (37)	5 (28)

N (%)

a) Adverse events with an incidence of ≥20% in the metreleptin groups for subcutaneous administration or continuous subcutaneous administration.
b) A significant digit for an incidence is as described in the clinical study report.

In the subcutaneous administration group, the incidence of any adverse event occurring from Week 4 through Week 24 in obese subjects was 74% (17 of 23 subjects), 100% (8 of 8 subjects), 100% (8 of 8 subjects), 83% (15 of 18 subjects), 83% (15 of 18 subjects), and 33% (1 of 3 subjects), respectively, in the placebo, 0.01, 0.03, 0.1, and 0.3 mg/kg (5 mg/mL formulation), and 0.3 mg/kg (20 mg/mL formulation) group, and the incidence of adverse drug reactions was 30% (7 of 23 subjects), 25% (2 of 8 subjects), 63% (5 of 8 subjects), 56% (10 of 18 subjects), 78% (14 of 18 subjects), and 33% (1 of 3 subjects), respectively. In the continuous subcutaneous administration group, the incidence of any adverse event occurring from Week 4 through Week 24 in obese subjects was 79% (15 of 19 subjects), 89% (16 of 18 subjects), 74% (14 of 19 subjects), and 61%

(11 of 18 subjects), in the placebo, and 0.3, 1.0, and 2.0 mg/kg (5 mg/mL formulation) group, respectively, and the incidence of adverse drug reactions was 32% (6 of 19 subjects), 83% (15 of 18 subjects), 74% (14 of 19 subjects), and 56% (10 of 18 subjects), respectively.

No deaths were reported. Four serious adverse events occurred in 1 subject in the 0.1 mg/kg group of 5 mg/mL formulation for subcutaneous administration (dyspnoea/nausea/fatigue/palpitations). All of them were mild in severity and their causal relationship to the study drug was denied. In the subcutaneous administration group, adverse events leading to discontinuation occurred in 1 subject in the 0.3 mg/kg group of 20 mg/mL formulation (injection site erythema/injection site inflammation/injection site pruritus) for nonobese subjects, 2 subjects in the 0.1 mg/kg group of 5 mg/mL formulation (injection site erythema/injection site ecchymosis/injection site reaction, palpitations; 1 subject each) and 3 subjects in the 0.3 mg/kg group of 20 mg/mL formulation (injection site oedema/injection site erythema/injection site inflammation/injection site pain/injection site pruritus, injection site erythema/injection site mass/injection site inflammation, injection site erythema/injection site inflammation/injection site pruritus/dermatitis contact; 1 subject each) for obese subjects (up to Week 4), and 1 subject in the 0.01 mg/kg group of 5 mg/mL formulation (injection site erythema/injection site pruritus/injection site rash/injection site urticaria) for obese subjects (from Week 4 through Week 24). In the continuous subcutaneous administration group, adverse events leading to discontinuation occurred in 1 subject in the 0.3 mg/kg group of 5 mg/mL formulation (malaise/headache/insomnia), 1 subject in the 1.0 mg/kg group of 5 mg/mL formulation (injection site oedema/injection site erythema/injection site mass/erythema/pruritus/urticaria), and 3 subjects in the 2.0 mg/kg group of 5 mg/mL formulation (diarrhoea/vomiting [1], injection site mass/injection site pain [2]) for nonobese subjects, 1 subject in the 0.3 mg/kg group of 5 mg/mL formulation (injection site mass), 2 subjects in the 1.0 mg/kg group of 5 mg/mL formulation (injection site mass, injection site erythema/injection site inflammation; 1 subject each), and 3 subjects in the 2.0 mg/kg group of 5 mg/mL formulation (injection site erythema/injection site pruritus/injection site inflammation, paraesthesia/pruritus/pyrexia, injection site mass; 1 subject each) for obese subjects (up to Week 4), and 1 subject in the 2.0 mg/kg group of 5 mg/mL formulation (injection site inflammation) for obese subjects (from Week 4 through Week 24). There were no clinically significant changes in laboratory tests, vital signs, or ECG.

The results of anti-leptin antibody measurement were as shown in Table 14.

Table 14. Results of anti-leptin antibody measurement

Dose (mg/kg/day)		Subcutaneous administration group							Continuous subcutaneous administration group				
		Placebo	5 mg/mL formulation				20 mg/mL formulation	Metreleptin group, total	Placebo	5 mg/mL formulation			Metreleptin group, total
			0.01	0.03	0.1	0.3	0.3			0.3	1.0	2.0	
Total number of subjects		42	16	16	31	26	7	96	31	27	27	27	81
Nonobese subjects[a)]	N	19	8	8	13	8	4	41	12	9	8	9	26
	N (measured)	15	8	8	13	8	2	39	12	8	6	5	19
	Positive	0	0	3	3	2	1	9	0	5	6	5	16
Obese subjects[b)]	N	23	8	8	18	18	3	55	19	18	19	18	55
	N (measured)	22	8	8	16	17	0	49	18	15	13	11	39
	Positive	1	4	5	13	15	0	37	1	13	13	10	36

a) The treatment period was 4 weeks.
b) The treatment period was 24 weeks.

4.(ii).A.(2) Studies in patients

4.(ii).A.(2).1) Japanese investigator-initiated trial (5.3.5.2-1, Study KUTR-003-1 [November 2010 to October 2011])

An open-label, uncontrolled study was conducted to evaluate the efficacy and safety of metreleptin in Japanese patients with lipodystrophy[55] (target sample size of 3).

All of 4 subjects enrolled were included in the safety analysis set and full analysis set (FAS)[56] [for the details of the study design, patient demographics, efficacy, and safety, see "4.(iii).A.(1) Japanese investigator-initiated trial"]. No subjects discontinued the trial.

For pharmacokinetics, diurnal variations in endogenous serum leptin concentrations before the start of treatment with metreleptin (within 7 days of the start of treatment) were as shown in Table 15.

Table 15. Diurnal variations in endogenous serum leptin concentrations before the start of treatment with metreleptin (within 7 days of the start of treatment)

Subject No. (sex/age)	0 hour	1 hour	2 hours	3 hours	4 hours	6 hours	9 hours	12 hours	16 hours	24 hours
K01 (female/18 years)	1.2	1.2	1.4	1.3	1.3	1.4	1.4	1.5	2.0	1.4
K02 (female/23 years)	4.8	3.5	4.6	3.3	3.0	3.7	3.4	3.9	4.0	3.6
K03 (female/11 years)	1.1	1.4	1.2	1.0	1.5	0.9	1.4	1.3	1.1	1.4
K04 (male/6 years)	1.1	1.9	1.5	0.9	1.5	1.3	1.3	1.5	1.0	1.4

Unit, ng/mL
The time represents the time elapsed from the start of blood collection for this evaluation.

Serum leptin concentrations over time following multiple subcutaneous administration of metreleptin for 20 weeks were as shown in Table 16.

[55] Major inclusion criteria: Patients aged ≥6 years with no apparent eating disorder who have diabetes mellitus (2010 Diagnostic Criteria for Diabetes Mellitus by the Japan Diabetes Mellitus Society) or hyperinsulinemia (fasting insulin > 30 μU/mL) associated with a decrease in adipose tissue/loss of adipose tissue and hypoleptinemia (< 3.0 ng/mL for males and < 6.0 ng/mL for females).

[56] Measurement of serum leptin concentrations is included in the efficacy endpoint. In addition to the measurement of trough concentrations, serum leptin concentrations were also determined at baseline and 1, 2, 3, 4, 6, 9, 12, 16, and 24 hours post-dose to evaluate diurnal variations in leptin.

Table 16. Serum leptin concentrations over time following multiple subcutaneous administration of metreleptin for 20 weeks

Subject No. (sex/age)	Dose[a] (mg/kg)	Serum leptin concentration (ng/mL)									
		Baseline	1 hour	2 hours	3 hours	4 hours	6 hours	9 hours	12 hours	16 hours	24 hours
K01 (female/ 18 years)	0.02 (Week 4)	1.7	3.8	4.6	5.1	6.5	4.1	4.4	3.9	2.8	2.2
	0.04 (Week 8)	2.9	9.8	11.5	12.5	12.0	10.9	11.3	7.1	6.5	2.6
	0.08 (Week 20)	9.4	94.7	120	123	127	134	102	75.1	19.1	10.5
K02 (female/ 23 years)	0.02 (Week 4)	3.3	13.5	19.8	18.6	18.6	12.6	8.2	6.0	4.0	3.8
	0.04 (Week 8)	11.9	75.3	133	119	119	87.7	38.5	24.3	15.2	9.8
	0.08 (Week 20)	97.0	169	208	197	241	202	177	130	97.7	69.9
K03 (female/ 11 years)	0.015 (Week 4)	1.1	5.1	6.3	4.6	3.0	2.7	2.0	2.0	1.3	1.3
	0.03 (Week 8)	2.1	11.4	18.4	16.7	10.8	6.9	4.8	3.0	2.6	1.4
	0.06 (Week 20)	8.3	28.9	37.8	70.4	67.9	33.3	25.3	17.0	11.3	7.8
K04 (male/6 years)	0.01 (Week 4)	1.6	3.2	2.5	2.5	1.8	1.6	1.4	1.0	1.5	1.2
	0.02 (Week 8)	6.6	10.7	13.5	14.2	12.2	11.8	8.3	7.7	6.5	6.2
	0.04 (Week 20)	19.3	25.2	35.5	34.3	30.5	36.4	25.4	21.2	18.3	19.7

a) Upper column, an estimated dose at which 50% of normal serum leptin concentration is achieved in each subject; middle column, an estimated dose at which 100% of normal serum leptin concentration is achieved in each subject; lower column, an estimated dose at which 200% of normal serum leptin concentration is achieved in each subject

Pharmacokinetic parameters following multiple subcutaneous administration of metreleptin for 20 weeks were as shown in Table 17.

Table 17. Pharmacokinetic parameters following multiple subcutaneous administration of metreleptin for 20 weeks

Subject No. (sex/age)	Dose[a] (mg/kg)	C_{max} (ng/mL)	t_{max} (h)	$AUC_{0-\tau}$ (ng·h/mL)	$t_{1/2,z}$ (h)	CL/F (L/h/kg)
K01 (female/ 18 years)	0.02 (Week 4)	6.5	3.8	84.98	14.7	0.235
	0.04 (Week 8)	12.5	2.8	181.8	7.47	0.220
	0.08 (Week 20)	134	6.0	1541	4.45	0.0519
K02 (female/ 23 years)	0.02 (Week 4)	19.8	1.8	193.9	8.07	0.103
	0.04 (Week 8)	133	1.8	1029	5.28	0.0389
	0.08 (Week 20)	241	3.9	3385	11.3	0.0236
K03 (female/ 11 years)	0.015 (Week 4)	6.3	1.9	53.56	16.1	0.280
	0.03 (Week 8)	18.4	2.0	125.2	11.5	0.240
	0.06 (Week 20)	70.4	2.8	546.7	8.43	0.110
K04 (male/6 years)	0.01 (Week 4)	3.2	1.1	37.23	17.9	0.269
	0.02 (Week 8)	14.2	2.9	204.9	17.2	0.0976
	0.04 (Week 20)	36.4	5.8	582.6	23.3	0.0687

C_{max}, maximum serum concentration; t_{max}, time to reach maximum serum concentration; $AUC_{0-\tau}$, area under the serum concentration-time curve within a dosing interval; $t_{1/2,Z}$, terminal elimination half-life; CL/F, apparent clearance

a) Upper column, an estimated dose at which 50% of normal serum leptin concentration is achieved in each subject; middle column, an estimated dose at which 100% of normal serum leptin concentration is achieved in each subject; lower column, an estimated dose at which 200% of normal serum leptin concentration is achieved in each subject

Serum leptin concentrations (trough concentrations) over time following multiple subcutaneous administration of metreleptin for 20 weeks were as shown in Table 18. At Week 20, which was the time point at which 200% of normal serum leptin concentration was estimated to be achieved in each subject, serum leptin concentrations (trough concentrations) in all 4 subjects reached not less than the mean endogenous serum leptin concentrations in Japanese healthy adult subjects[57] (3.9 ng/mL for male subjects and 7.3 ng/mL for female subjects).

[57] Ogawa T, et al., *Metabolism*. 2004;53(7):879-885. Results were obtained from Japanese healthy adult male subjects (n = 198; mean age, 44.5 years; mean BMI, 22.9 kg/m^2) and Japanese healthy adult female subjects (n = 221; mean age, 41.7 years; mean BMI, 20.6 kg/m^2).

Table 18. Serum leptin concentrations (trough concentrations) over time following multiple subcutaneous administration of metreleptin for 20 weeks

	Timepoint	Baseline	Week 4	Week 8	Week 20
	Dose[a)]	-	0.01[b)] 0.015[b)] 0.02[b)]	0.02[c)] 0.03[c)] 0.04[c)]	0.04[d)] 0.06[d)] 0.08[d)]
Subject No. (sex/age)	K01 (female/18 years)	1.2	1.7	2.9	9.4
	K02 (female/23 years)	4.8	3.3	11.9	97.0
	K03 (female/11 years)	1.1	1.1	2.1	8.3
	K04 (male/6 years)	1.1	1.6	6.6	19.3

Unit, ng/mL

a) Upper column, male subjects; middle column, female subjects (aged <18 years); lower column, female subjects (aged≥18 years)
b) An estimated dose at which 50% of normal serum leptin concentration is achieved in each subject
c) An estimated dose at which 100% of normal serum leptin concentration is achieved in each subject
d) An estimated dose at which 200% of normal serum leptin concentration is achieved in each subject

4.(ii).A.(2).2) Japanese clinical research (Reference data 5.3.5.2-2, Study KUTR-003-0 [May 2002 to August 2011, data cut-off date])

An open-label, uncontrolled study was conducted to evaluate the efficacy and safety of metreleptin in Japanese patients with lipodystrophy.[58]

Of 12 subjects enrolled, 11 subjects excluding 1 subject who withdrew consent, were included in the efficacy evaluable population[59] and 9 subjects excluding 2 subjects who discontinued the study (physician's decision, adverse events; 1 subject each) completed 1-year treatment. Eight subjects excluding 1 subject who discontinued treatment from year 2 to the data cut-off date due to surgery for bile duct carcinoma[60], continued to receive treatment [for the details of the study design, patient demographics, efficacy, and safety, see "4.(iii).A.(2) Japanese clinical research"].

For pharmacokinetics, trough concentrations over time following multiple subcutaneous administration of metreleptin for 1 year were as shown in Table 19. At Month 4, which was the time point at which 200% of normal serum leptin concentration was estimated to be achieved in each subject, serum leptin concentrations (trough concentrations) in 9 of 10 subjects reached not less than the mean of endogenous serum leptin concentrations in Japanese healthy adult subjects[57] (3.9 ng/mL for male subjects and 7.3 ng/mL for female subjects).

[58] Major inclusion criteria: Patients aged ≥5 years who have diabetes mellitus (plasma glucose level on an oral glucose tolerance test [OGTT] > 200 mg/dL, HbA1c ≥ 6.5% [NGSP value]), hyperinsulinemia (fasting insulin > 30 μU/mL), or hypertriglyceridemia (fasting triglyceride > 200 mg/dL) associated with obvious fat atrophy and hypoleptinemia (< 3.0 ng/mL for males and < 6.0 ng/mL for females).

[59] Measurement of serum leptin concentrations is included in the efficacy endpoint.

[60] Since the patient had a routine checkup in the second year of leptin treatment and abdominal CT scan findings were suggestive of liver cancer, leptin treatment was discontinued and tumor resection was performed. The patient was given a pathological diagnosis of cholangiocellular carcinoma during the surgery. Lymph node metastasis was observed at this point and the patient underwent radiotherapy and chemotherapy after the surgery; however, the patient's condition worsened and died at 8 months after the surgery.

Table 19. Serum leptin concentrations (trough concentrations) over time following multiple subcutaneous administration of metreleptin for 1 year

	Timepoint	Baseline	At year 1					
			Month 1	Month 2	Month 4	Month 6	Month 8	Month 12
	Dose[a] (mg/kg)	-	0.01[b] 0.015[b] 0.02[b]	0.02[c] 0.03[c] 0.04[c]	0.04[d] 0.06[d] 0.08[d]			
Subject No. (sex/age)	01 (female/ 11 years)	0.94	1.51	4.02	1.29	0.78	1.35	2.12
	02 (male/ 29 years)	0.82	4.15	3.96	21.69	25.28	41.18	69.28
	03 (male/ 19 years)	1.22	ND	3.36	28.67	32.63	34.63	43.57
	04 (female/ 16 years)	1.15	2.17	3.32	9.40	3.81	27.52	50.58
	05 (female/ 22 years)	1.41	1.48	2.09	8.56	-	-	-
	06 (female/ 15 years)	1.02	1.44	3.54	30.03	61.44	28.31	78.82
	08 (female/ 33 years)	1.40	4.16	11.09	-	-	-	-
	09 ([redacted] years)	1.24	1.92	12.35	44.85	27.36	24.77	80.44
	10 ([redacted] years)	0.49	1.65	6.88	93.55	111.15	149.55	76.83
	11 ([redacted] years)	0.70	0.06	2.51	20.15	50.15	45.65	ND
	12 ([redacted] years)	1.50	ND	1.18	18.80	44.85	79.45	82.20
N		11	9	11	10	9	9	8
Mean ± SE		1.08 ± 0.10	2.06 ± 0.44	4.94 ± 1.10	27.70 ± 8.31	39.72 ± 11.14	48.05 ± 14.46	60.48 ± 9.77

Unit, ng/mL; -, not applicable; ND, not detected (missing data)

a) Upper column, male subjects; middle column, female subjects (aged <18 years); lower column, female subjects (aged ≥18 years)
b) An estimated dose at which 50% of normal serum leptin concentration is achieved in each subject
c) An estimated dose at which 100% of normal serum leptin concentration is achieved in each subject
d) An estimated dose at which 200% of normal serum leptin concentration is achieved in each subject

Serum leptin concentrations (trough concentrations) over time following multiple subcutaneous administration of metreleptin from year 2 to year 9 were as shown in Table 20.

Table 20. Serum leptin concentrations (trough concentrations) over time following multiple subcutaneous administration of metreleptin from year 2 to year 9[a]

Subject No. (sex/age)	At year 2 Month 12	At year 3 Month 12	At year 4 Month 12	At year 5 Month 12	At year 6 Month 12	At year 7 Month 12	At year 8 Month 12	At year 9 Month 12
01 (female/11 years)	ND	1.01	0.38	9.37	0.52	0.59	ND	3.64
02 (male/29 years)	59.95	36.83	11.29	6.63	3.07	15.03	11.30	-
03 (male/19 years)	8.66	1.96	1.83	2.72	1.48	4.07	ND	-
04 (female/16 years)	1.43	1.43	ND	2.63	1.63	2.09	-	-
06 (female/15 years)	12.19	ND	3.98	9.62	4.21	8.20	-	-
09 ([redacted] years)	10.05	1.60	7.00	19.90	-	-	-	-
10 ([redacted] years)	64.27	-	-	-	-	-	-	-
11 ([redacted] years)	24.50	-	-	-	-	-	-	-
12 ([redacted] years)	-	-	-	-	-	-	-	-
N	7	5	5	6	5	5	1	1
Mean ± SE	25.86 ± 9.72	8.57 ± 7.07	4.90 ± 1.95	8.48 ± 2.60	2.18 ± 0.65	6.00 ± 2.60	11.30	3.64

Unit, ng/mL; -, not applicable; ND, not detected (missing data)

a) The total daily dose at year 1 was not changed, but the dosing frequency was changed from twice daily to once daily.

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

4.(ii).B.(1) Effect of unphysiologically high serum leptin concentrations on safety

PMDA asked the applicant to explain the effect of unphysiologically high serum leptin concentrations associated with the treatment with metreleptin on safety.

The applicant responded as follows:

The mean serum leptin concentrations in Japanese healthy adult subjects are 3.9 ng/mL for male and 7.3 ng/mL for female.[57] Of the 4 subjects who had participated in the Japanese investigator-initiated trial (Study KUTR-003-1), 1 subject (Subject No. K02) had a serum leptin concentration (trough concentration) of 97.0 ng/mL on Day 139 of treatment with metreleptin and the C_{max} was also observed in the subject on the same day. No new adverse event was reported for the treatment period. Of the 12 subjects enrolled in the Japanese clinical research (Study KUTR-003-0), 1 subject (Subject No. 10) had a serum leptin concentration (trough concentration) of 149.55 ng/mL on Day 251 of treatment. Only palpitations (which occurred on Day 247 and resolved on Day 254) was reported as an adverse event for the treatment period. For 6 subjects who had serum leptin concentrations (trough concentrations) ≥50 ng/mL at year 1 in the Japanese clinical research (Study KUTR-003-0) (Subject Nos. 02, 04, 06, 09, 10, and 12), the adverse events reported around 12 months after the start of treatment were eczema (mild; it occurred on Day 394 of treatment and resolved on Day 470) in Subject No. 02, abdominal pain and oropharyngeal pain (mild; it occurred on Day 410 and resolved on Day 411) in Subject No. 04, abdominal pain (mild; it occurred on Day 326 and resolved on Day 337), diarrhoea (mild; it occurred on Day 330 and resolved on Day 337), and nausea (mild; it occurred on Day 388 and resolved on Day 437) in Subject No. 06, blood pressure increased (mild; it occurred on Day 363 and persisted), varicose vein (mild; it occurred on Day 364 and persisted), and headache (mild; it occurred on Day 372 and resolved on Day 376) in Subject No. 10, and no adverse events were reported in Subject No. 09 or 12 during this period.

The above results have indicated that no specific adverse events tend to occur in high serum leptin concentrations. Anorexia due to leptin-induced appetite suppression has not been reported and there is no clear tendency of occurrence for elevated blood pressure, which is the physiological action of leptin, in the Japanese studies. Therefore, high serum leptin concentrations are unlikely to cause specific adverse events. The applicant intends to continuously evaluate the safety of long-term exposure to high serum leptin concentrations.

PMDA considers as follows:

PMDA accepts the applicant's response regarding the short-term effect of unphysiologically high serum leptin concentrations on safety, but the long-term effect of metreleptin on safety is unknown. In clinical practice, the measurement of serum leptin concentrations is not routinely performed and therefore, it cannot be ruled out that high serum leptin concentrations may persist for a long time. On the other hand, in the foreign phase I studies in foreign healthy adult subjects (Studies LEPT-[redacted]0121 and LEPT-[redacted]0272), subjects received higher doses of metreleptin (for the maximum dose, 3.0 mg/kg of 5 mg/mL formulation in the foreign phase I intravenous study [Study LEPT-[redacted]0121], 0.3 mg/kg of 5 mg/mL formulation in the foreign phase I subcutaneous study [Study LEPT-[redacted]0272], and 0.08 mg/kg of 5 mg/mL formulation in the Japanese investigator-initiated trial [Study KUTR-003-1]) for about 4 to 24 weeks when compared with those participating in Japanese studies such as the Japanese investigator-initiated trial (Study KUTR-003-1). The subjects were considered to might have been continuously exposed to high levels of metreleptin (the mean

serum leptin concentrations following administration of 3.0 mg/kg [5 mg/mL formulation] was 1370.7 ng/mL on Day 15 in the foreign phase I subcutaneous administration study [Study LEPT-[redacted]0121]), but no tendency for a high rate of occurrence of specific adverse events has been observed. The above results indicate that unphysiologically high serum leptin concentrations have not raised specific concerns on long-term safety, but more information needs to be collected via post-marketing surveillance.

4.(ii).B.(2) Effect of antibodies on pharmacokinetics and efficacy

PMDA asked the applicant to explain the effect of antibodies on pharmacokinetics and efficacy.

The applicant responded as follows:

With respect to the effect of antibody expression on pharmacokinetics, as anti-leptin antibodies were produced, the tendency for the increase in serum leptin concentration was observed in the foreign phase I studies (Studies LEPT-[redacted]0121 and LEPT-[redacted]0272).[61] The production of anti-leptin antibody may have affected the level of leptin exposure to increase, and had an effect on the measurements of serum leptin concentrations. As for the effect of antibody production on efficacy, the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769) reported that metreleptin-binding antibodies had been produced in 60% (12 of 20 subjects) of lipodystrophy patients treated with metreleptin, and the effects on HbA1c and triglyceride, both of which were the efficacy endpoints, were examined for the antibody-negative group (n = 8) and antibody-positive group (n = 12). As a result, HbA1c levels decreased from 8.29% at baseline to 7.37% at 12 months after the start of treatment in the antibody-negative group and from 9.22% to 7.30% in the antibody-positive group, and there were no relevant differences between the two groups. At 12 months after the start of treatment, triglyceride levels decreased from 532 mg/dL at baseline to 162.8 mg/dL in the antibody-negative group and from 1890.1 mg/dL to 645.7 mg/dL in the antibody-positive group. There were no major differences in the rate of decrease between the two groups. The above results indicate that the antibody expression after the treatment with metreleptin does not affect efficacy.

PMDA considers as follows:

The applicant's explanation from the perspective of pharmacokinetics is acceptable, but it is necessary to continue to examine the effects of antibody production on efficacy and safety in Clinical Section ["4.(iii).B.(2).3) Effect of antibodies" and "4.(iii).B.(3).5) Effect of antibodies"].

4.(iii) Summary of clinical efficacy and safety

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

As the evaluation data, the results from a Japanese investigator-initiated trial (Study KUTR-003-1) were submitted. As the reference data, the results from a Japanese clinical research (Study KUTR-003-0), a Japanese investigational study (Study KUTR-003-2)[46], a foreign NIH clinical study (Study

[61] In the foreign phase I intravenous study (Study LEPT-[redacted]0121), the median serum leptin concentrations following multiple intravenous administration of 0.3 mg/kg (frozen formulation) ranged from 2.04 to 3.74 ng/mL and from 9.16 to 63.5 ng/mL, respectively, in the group where anti-leptin antibodies were not produced until 8 days after completion of treatment and the group where anti-leptin antibodies were produced. In the foreign phase I subcutaneous study (Study LEPT-[redacted]0272), the median serum leptin concentrations (trough concentrations) following single subcutaneous administration of 0.3 mg/kg (frozen formulation) ranged from 4.16 to 18.64 ng/mL and from 16.39 to 81.75 ng/mL, respectively, in the group where anti-leptin antibodies were not produced and the group where anti-leptin antibodies were produced.

[redacted]1265/20[redacted]0769), and a foreign Amylin-sponsored study (Study FHA101) were submitted (for subject transfer in the Japanese clinical studies, see Figure 1). The results from the main studies are described below. In the Japanese studies, HbA1c is expressed as a JDS value. The applicant has defined 100% dose as a dosage in which is expected to raise the serum leptin concentration to the levels in humans with 20% (males) or 30% (females) body fat, which are the upper limit of the normal range of body fat percentage, and daily dose as an estimated dose at which 50%, 100%, and 200% of normal serum leptin concentration[62] are achieved (hereinafter referred to as "50% dose," "100% dose," and "200% dose").

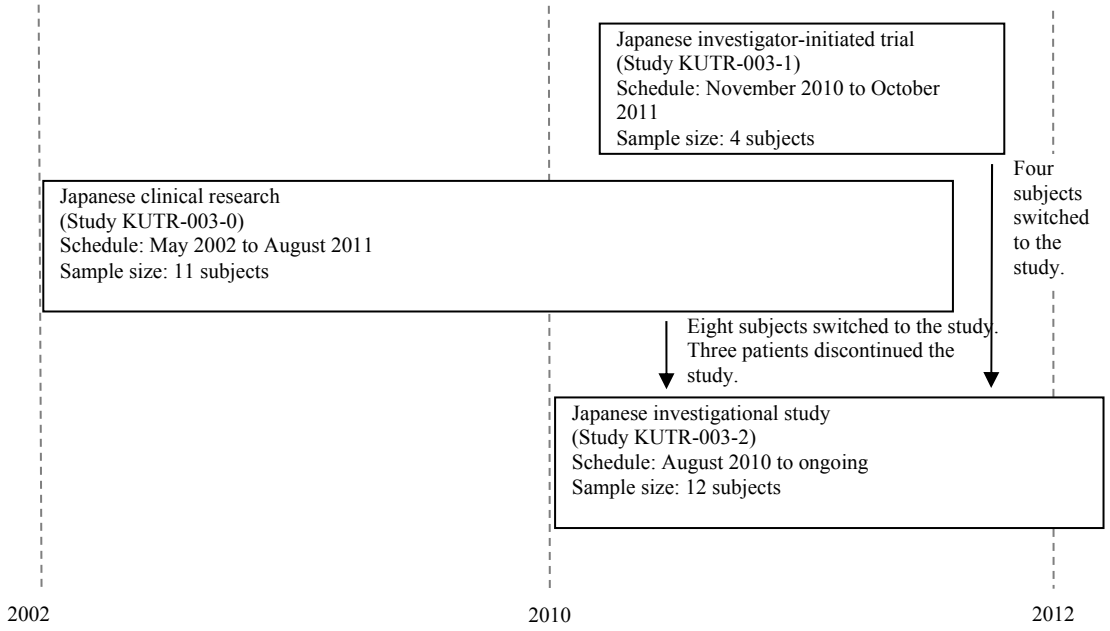

Figure 1. Subject transfer in the Japanese clinical studies

4.(iii).A.(1) Japanese investigator-initiated trial **(5.3.5.2-1, Study KUTR-003-1 [November 2010 to October 2011])**

An open-label, uncontrolled study was conducted to evaluate the efficacy and safety of metreleptin in Japanese patients with lipodystrophy[55] (target sample size of 3).

In male subjects, metreleptin 0.01 mg/kg (50% dose) was to be administered subcutaneously once daily for the first 4 weeks, followed by 0.02 mg/kg (100% dose) from Week 5 to Week 8, and then by 0.04 mg/kg (200% dose) from Week 9 onwards. In Female subjects aged <18 years, metreleptin 0.015 mg/kg (50% dose) was to be administered subcutaneously once daily for the first 4 weeks, followed by 0.03 mg/kg (100% dose) from Week 5 to Week 8, and then by 0.06 mg/kg (200% dose) from Week 9 onwards. In Female subjects aged ≥18 years, metreleptin 0.02 mg/kg (50% dose) was administered subcutaneously once daily for the first 4 weeks, followed by 0.04 mg/kg (100% dose) from Week 5 to Week 8, and then by 0.08 mg/kg (200% dose) from Week 9 onwards. The treatment period was 20 weeks. When subjects received concomitant medications

62 The mean endogenous leptin concentration is calculated to be 6.14 ng/mL in a male subject with a body weight of 80 kg and a body fat percentage of 20% according to the formula "$\log_{10}$[mean endogenous leptin concentration (ng/mL)] = 0.34 + 0.028 × amount of fat (kg)," and it is calculated to be 15.8 ng/mL in a female subject with a body weight of 60 kg and a body fat percentage of 30% according to the formula "$\log_{10}$[mean endogenous leptin concentration (ng/mL)] = 0.75 + 0.025 × amount of fat (kg)."

for the treatment of diabetes mellitus or dyslipidemia, dose reduction or discontinuation of the concomitant medications were allowed as appropriate. Patients injected themselves with the drug product.

All of 4 subjects enrolled were included in the safety analysis set and FAS. Subject demographics (at the time of enrollment) were as shown in Table 21.

Table 21. Subject demographics (at the time of enrollment)

Subject No. (sex/age)	Classification	Body weight (kg)	BMI (kg/m^2)	Complication		
				Diabetes mellitus	Hyperinsulinemia	Hypertriglyceridemia
K01 (female/ 18 years)	Congenital generalized	[redacted]	17.24	Yes	Yes	Yes
K02 (female/ 23 years)	Acquired partial	[redacted]	16.85	Yes	No	Yes
K03 (female/ 11 years)	Congenital generalized	[redacted]	17.8	Yes	Yes	Yes
K04 (male/ 6 years)	Congenital generalized	[redacted]	14.63	No	Yes	Yes

For efficacy, a mean change from baseline in HbA1c at Week 20, the primary endpoint, and its 95% CI was −1.53 (−4.02, 0.97). HbA1c levels over time in each subject were as shown in Table 22.

Table 22. HbA1c levels over time in each subject

Subject No. (sex/age)	Baseline	Week 4 (Month 1)	Week 8 (Month 2)	Week 12 (Month 3)	Week 16 (Month 4)	Week 20 (Month 5)
	-	50% dose	100% dose	200% dose		
K01 (female/18 years)[a)]	8.6	7.7	6.5	5.5	4.8	4.8
K02 (female/23 years)[b)]	7.7	6.2	5.7	5.6	5.9	6.4
K03 (female/11 years)[c)]	5.8	5.3	5.2	5.1	5.1	5.4
K04 (male/6 years)	5.8	5.2	5.1	5.0	5.1	5.2

Unit, %; -, not applicable

a) Pioglitazone hydrochloride (30 mg/day), voglibose (0.6 mg/day), and metformin hydrochloride (500 mg/day) had been administered concomitantly since before the start of treatment and they were discontinued 4, 11, and 25 days after the start of treatment, respectively.

b) Insulin preparation (insulin aspart [genetical recombination] [10-75 units/day], insulin glargine [genetical recombination] [5-80 units/day]) had been administered concomitantly since before the start of treatment and they were discontinued 28 days after the start of treatment. Metformin hydrochloride (750-1250 mg/day) had been used concomitantly before the start of treatment and it was administered throughout the study period.

c) Voglibose (0.3 mg/day) and metformin hydrochloride (625 mg/day) had been administered concomitantly since before the start of treatment and they were discontinued 14 and 34 days after the start of treatment, respectively.

The parameters over time for the secondary endpoints were as shown in Table 23.

Table 23. Parameters over time for the secondary endpoints

Subject No. (sex/age)	Baseline	Week 4 (Month 1)	Week 8 (Month 2)	Week 12 (Month 3)	Week 16 (Month 4)	Week 20 (Month 5)
	-	50% dose	100% dose	200% dose		
Fasting blood glucose level (mg/dL)						
K01 (female/18 years)	115	82	80	67	85	77
K02 (female/23 years)	78	92	84	93	125	140
K03 (female/11 years)	77	72	91	80	85	73
K04 (male/6 years)	63	66	78	131	86	79
Fasting insulin (μU/mL)						
K01 (female/18 years)	10.8	2.8	10.6	2.3	4.3	7.7
K02 (female/23 years)	22.1	16.9	8.4	7.1	17.2	14.1
K03 (female/11 years)	7.2	5.1	14.5	1.3	7.2	9.5
K04 (male/6 years)	37.6	ND	ND	ND	18.0	20.9
Triglyceride (mg/dL)						
K01 (female/18 years)[a)]	210	144	78	55	55	62
K02 (female/23 years)[b)]	246	161	98	51	144	204
K03 (female/11 years)	59	49	52	46	60	77
K04 (male/6 years)	180	163	88	83	131	382

Unit, mg/dL; -, not applicable; ND, not detected (missing data); See the footnote of Table 22 for usage of concomitant antidiabetic drugs.
a) Bezafibrate (400 mg/day) had been administered concomitantly since before the start of treatment and it was discontinued 45 days after the start of treatment.
b) Bezafibrate (200-400 mg/day) had been administered concomitantly since before the start of treatment and it was discontinued 40 days after the start of treatment.

For safety, the occurrence of adverse events and adverse drug reactions was as shown in Table 24.

Table 24. Occurrence of adverse events and adverse drug reactions (safety analysis set, 4 subjects)

Name of the event	Adverse event	Adverse drug reaction
Total	4 (100.0)	4 (100.0)
Dry skin	3 (75.0)	2 (50.0)
Headache	2 (50.0)	2 (50.0)
Alopecia	2 (50.0)	2 (50.0)
Hepatic enzyme increased	2 (50.0)	1 (25.0)
Epistaxis	2 (50.0)	1 (25.0)
Wrong technique in drug usage process	1 (25.0)	1 (25.0)
Hypoglycaemia	1 (25.0)	1 (25.0)
Rash	1 (25.0)	1 (25.0)
Constipation	1 (25.0)	1 (25.0)
Pruritus	1 (25.0)	0 (0.0)
Influenza	1 (25.0)	0 (0.0)
Hyperkeratosis	1 (25.0)	0 (0.0)
Intentional self-injury	1 (25.0)	0 (0.0)
Excoriation	1 (25.0)	0 (0.0)
Toothache	1 (25.0)	0 (0.0)
Auricular haematoma	1 (25.0)	0 (0.0)
Upper respiratory tract infection	1 (25.0)	0 (0.0)
Albumin urine present	1 (25.0)	0 (0.0)
Insomnia	1 (25.0)	0 (0.0)

N (%); MedDRA/J ver. 15.0

No deaths, serious adverse events, or adverse events leading to discontinuation were reported.

The body weight measured at baseline and Week 20 was [redacted] kg and [redacted] kg, respectively, in Subject No. K01, [redacted] kg and [redacted] kg, respectively, in K02, [redacted] kg and [redacted] kg, respectively, in K03, and [redacted] kg and [redacted] kg, respectively, in K04.

4.(iii).A.(2) Japanese clinical research (Reference data 5.3.5.2-2, Study KUTR-003-0 [May 2002 to August 2011, data cut-off date])

An open-label, uncontrolled study was conducted to evaluate the efficacy and safety of metreleptin in Japanese patients with lipodystrophy.[58]

In male subjects, metreleptin 0.01 mg/kg (50% dose) was to be administered subcutaneously twice daily for the first 1 month, followed by 0.02 mg/kg (100% dose) during Month 2, and then by 0.04 mg/kg (200% dose) from Month 3 to Month 12. In female subjects aged <18 years, 0.015 mg/kg (50% dose) were to be administered subcutaneously twice daily for the first 1 month, followed by 0.03 mg/kg (100% dose) during Month 2, and then by 0.06 mg/kg (200% dose) from Month 3 to Month 12. In female subjects aged ≥18 years, 0.02 mg/kg (50% dose) was to be administered subcutaneously twice daily for the first 1 month, followed by 0.04 mg/kg (100% dose) during Month 2, and then by 0.08 mg/kg (200% dose) from Month 3 to Month 12. After Month 12, the dosing frequency was changed from twice daily to once daily while the total daily dose was maintained. When subjects received concomitant medications for the treatment of diabetes mellitus or dyslipidemia, dose reduction or discontinuation of the concomitant medications were allowed as appropriate.

Of the 12 subjects enrolled, 11 subjects, excluding 1 subject who withdrew consent, were included in the efficacy analysis set, and 9 subjects, excluding 2 subjects who discontinued the study (physician's decision, adverse events; 1 subject each), were considered to have completed 1-year treatment. Eight subjects, excluding 1 subject who discontinued treatment from Year 2 to the data cut-off date due to surgery for bile duct carcinoma[60], were considered to continue to receive treatment. Subject demographics (at the time of enrollment) were as shown in Table 25.

Table 25. Subject demographics (at the time of enrollment)

Subject No. (sex/age)	Classification	Body weight (kg)	BMI (kg/m^2)	Complication		
				Diabetes mellitus	Hyperinsulinemia	Hypertriglyceridemia
01 (female/11 years)	Acquired generalized	30	15.48	Yes	Yes	Yes
02 (male/29 years)	Congenital generalized	32	14.2	Yes	No	No
03 (male/19 years)	Congenital generalized	52.8	19.68	Yes	No	No
04 (female/16 years)	Acquired generalized	33.2	13.37	Yes	No	Yes
05 (female/22 years)	Congenital generalized	52	21.2	Yes	No	Yes
06 (female/15 years)	Congenital generalized	44	18.22	Yes	No	No
08 (female/33 years)	Congenital generalized	53.9	20.31	Yes	No	No
09 ([redacted] years)	Congenital generalized	[redacted]	18.73	Yes	No	Yes
10 ([redacted] years)	Congenital generalized	[redacted]	18.28	Yes	Yes	No
11 ([redacted] years)	Congenital generalized	[redacted]	24.69	No	No	Yes
12 ([redacted] years)	Familial partial	[redacted]	18.8	Yes	No	No

For efficacy, HbA1c, fasting blood glucose, fasting insulin, and triglyceride levels over time for 12 months after the start of treatment with metreleptin were as shown in Table 26.

Table 26. Mean HbA1c, fasting blood glucose, fasting insulin, and triglyceride levels over time for 12 months after the start of treatment with metreleptin

	Baseline	Year 1					
		Month 1	Month 2	Month 4	Month 6	Month 8	Month 12
Dose[a] (mg/kg)	-	0.01[b] 0.015[b] 0.02[b]	0.02[c] 0.03[c] 0.04[c]	0.04[d] 0.06[d] 0.08[d]			
HbA1c (%)	8.74 ± 0.62 (11)	6.47 ± 0.24 (11)	5.85 ± 0.23 (10)	6.01 ± 0.41 (10)	5.84 ± 0.21 (9)	5.89 ± 0.24 (9)	5.83 ± 0.31 (9)
Fasting blood glucose level (mg/dL)	137.5 ± 15.8 (11)	113.3 ± 10.1 (11)	105.5 ± 12.7 (11)	95.7 ± 8.6 (10)	103.9 ± 11.7 (9)	93.3 ± 4.9 (9)	89.0 ± 2.0 (9)
Fasting insulin (μU/mL)	18.13 ± 4.97 (11)	13.18 ± 3.21 (11)	12.77 ± 3.51 (11)	25.33 ± 13.99 (10)	7.61 ± 1.19 (9)	7.33 ± 1.44 (9)	7.84 ± 2.40 (9)
Triglyceride (mg/dL)	395.4 ± 185.9 (11)	164.0 ± 49.5 (11)	105.5 ± 25.1 (11)	102.2 ± 16.4 (10)	103.9 ± 20.2 (9)	100.4 ± 21.9 (9)	88.7 ± 13.4 (9)

Mean ± SE (N)

a) Upper column, male subjects; middle column, female subjects (<18 years); lower column, female subjects (≥18 years)
b) An estimated dose at which 50% of normal serum leptin concentration is achieved in each subject
c) An estimated dose at which 100% of normal serum leptin concentration is achieved in each subject
d) An estimated dose at which 200% of normal serum leptin concentration is achieved in each subject

HbA1c, fasting blood glucose, fasting insulin, and triglyceride levels over time from 12 months after the start of treatment with metreleptin to data cut-off were as shown in Table 27.

Table 27. HbA1c, fasting blood glucose, fasting insulin, and triglyceride levels over time from 12 months after the start of treatment with metreleptin to data cut-off

	Year 2	Year 3	Year 4	Year 5	Year 6	Year 7	Year 8	Year 9
	Month 12	Month 12	Month 12	Month 12	Month 12	Month 12	Month 12	Month 12
Dose[a] (mg/kg)	0.04[b] 0.06[b] 0.08[b]							
HbA1c (%)	6.25 ± 0.57 (8)	6.17 ± 0.76 (6)	7.48 ± 1.03 (5)	7.22 ± 1.10 (6)	8.14 ± 1.57 (5)	7.62 ± 1.31 (5)	6.2, 11.9 (2)	5.70 (1)
Fasting blood glucose level (mg/dL)	95.0 ± 6.8 (8)	96.8 ± 5.9 (6)	110.6 ± 9.8 (5)	102.2 ± 10.2 (6)	131.2 ± 33.7 (5)	141.8 ± 34.6 (5)	83, 295 (2)	88.0 (1)
Fasting insulin (μU/mL)	9.46 ± 2.78 (8)	6.28 ± 2.11 (6)	5.44 ± 1.15 (5)	5.60 ± 1.67 (6)	7.54 ± 2.48 (5)	5.54 ± 1.95 (5)	12.1, 4.2 (2)	11.70 (1)
Triglyceride (mg/dL)	105.4 ± 32.1 (7)	79.5 ± 19.8 (6)	58.2 ± 7.3 (5)	77.6 ± 12.6 (5)	136.6 ± 54.5 (5)	135.4 ± 29.8 (5)	87, 161 (2)	106.0 (1)

Mean ± SE (N)

a) Upper column, male subjects; middle column, female subjects (<18 years); lower column, female subjects (≥18 years)
b) An estimated dose at which 200% of normal serum leptin concentration is achieved in each subject

For safety, adverse events and adverse drug reactions reported by at least 2 subjects were as shown in Table 28.

Table 28. Adverse events and adverse drug reactions occurring in at least 2 subjects (safety analysis set)

Name of the event	Adverse event	Adverse drug reaction
Total	11 (100)	6 (54.5)
Injection site reaction	7 (63.6)	6 (54.5)
Headache	7 (63.6)	0 (0)
Abdominal pain	5 (45.5)	0 (0)
Pyrexia	5 (45.5)	0 (0)
Malaise	5 (45.5)	0 (0)
Oropharyngeal pain	4 (36.4)	0 (0)
Diarrhoea	4 (36.4)	0 (0)
Nausea	4 (36.4)	0 (0)
Rash	4 (36.4)	0 (0)
Cough	4 (36.4)	0 (0)
Upper respiratory tract inflammation	4 (36.4)	0 (0)
Nasopharyngitis	3 (27.3)	0 (0)
Pharyngitis	3 (27.3)	0 (0)
Gastritis	3 (27.3)	0 (0)
Influenza	3 (27.3)	0 (0)
Palpitations	3 (27.3)	0 (0)
Gastroenteritis	2 (18.2)	0 (0)
Anaemia	2 (18.2)	0 (0)
Rhinorrhoea	2 (18.2)	0 (0)
Blood glucose decreased	2 (18.2)	0 (0)
Pain in extremity	2 (18.2)	0 (0)
Abdominal pain upper	2 (18.2)	0 (0)
Acne	2 (18.2)	0 (0)
Constipation	2 (18.2)	0 (0)
Corneal erosion	2 (18.2)	0 (0)
Arthropod sting	2 (18.2)	0 (0)
Conjunctivitis allergic	2 (18.2)	0 (0)
Eczema	2 (18.2)	0 (0)
Enterocolitis	2 (18.2)	0 (0)
Hypoaesthesia	2 (18.2)	0 (0)
Injury	2 (18.2)	0 (0)
Oral pain	2 (18.2)	0 (0)
Rhinitis allergic	2 (18.2)	0 (0)
Tinea pedis	2 (18.2)	0 (0)
Xeroderma	2 (18.2)	0 (0)
Hepatic mass	2 (18.2)	0 (0)

N (%); MedDRA/J ver. 14.0

Although no deaths or serious adverse events occurred during the study period, 1 subject who had discontinued treatment due to surgery for bile duct carcinoma[60] died after the discontinuation of treatment.

Adverse events leading to discontinuation occurred in 1 subject (leucopenia/white blood cell count decreased). The body weight (mean ± SE) measured before the start of treatment with metreleptin and at 12 months after the start of treatment was 46.51 ± 3.82 kg and 41.64 ± 3.39 kg, respectively.

4.(iii).A.(3) Japanese investigational study (Reference data 5.3.5.2-3, Study KUTR-003-2 [December 2010 to ongoing])

An open-label, uncontrolled study was conducted to evaluate the long-term safety and efficacy of metreleptin in Japanese patients with lipodystrophy who had received leptin replacement therapy.[63] The data obtained up to September 30, 2012 were summarized and submitted in this application.

[63] Major inclusion criteria: Patients aged ≥6 years who present with decreased adipose tissue or loss of adipose tissue, with no obvious eating

In male subjects, 0.04 mg/kg (200% dose) was to be administered subcutaneously once daily. In female subjects aged <18 years, 0.06 mg/kg (200% dose) was to be administered subcutaneously once daily, and in female subjects aged ≥18 years, 0.08 mg/kg (200% dose) was to be administered subcutaneously once daily. When the dose administered in preceding studies (Japanese investigator-initiated trial [Study KUTR-003-1] and Japanese clinical research [Study KUTR-003-0]) was <200% dose, subjects were to continue to receive the same dose and they were allowed to be treated with a twice-daily regimen if their physician considered it appropriate. When patients received concomitant medications for the treatment of diabetes mellitus or dyslipidemia, dose reduction and discontinuation of the concomitant medications were allowed as appropriate. Subjects injected themselves with the drug product.

All of the 12 subjects enrolled were included in the efficacy analysis set and safety analysis set. Of them, 4 and 8 subjects switched from the Japanese investigator-initiated trial (Study KUTR-003-1) and Japanese clinical research (Study KUTR-003-0), respectively, to this study.

For efficacy, HbA1c, fasting blood glucose, fasting insulin, and triglyceride levels over time before and after the switch to this study were as shown in Table 29.

Table 29. HbA1c, fasting blood glucose, fasting insulin, and triglyceride levels over time before and after the switch to this study

Endpoint	Before the switch to this study	At the time of cut-off in December 2011	At the time of cut-off in September 2012
HbA1c (%)	5.77 ± 1.21 (12)	6.00 ± 1.42 (12)	6.46 ± 2.24 (12)
Fasting blood glucose level (mg/dL)	95.50 ± 21.62 (12)	118.17 ± 54.53 (12)	127.00 ± 116.23 (12)
Fasting insulin (μU/mL)	12.75 ± 14.5 (12)	16.39 ± 18.52 (12)	8.77 ± 5.17 (12)
Triglyceride (mg/dL)	128.25 ± 101 (12)	128.83 ± 72.16 (12)	150.33 ± 93.82 (12)

Mean ± SD (N)

For safety, adverse events and adverse drug reactions reported by at least 2 subjects were as shown in Table 30.

Table 30. Adverse events and adverse drug reactions reported by at least 2 subjects (safety analysis set)

Name of the event	Adverse event	Adverse drug reaction
Total	11 (91.7)	8 (66.7)
Nasopharyngitis	6 (50.0)	1 (8.3)
Influenza	2 (16.7)	0 (0)
Conjunctivitis allergic	2 (16.7)	0 (0)
Acne	2 (16.7)	2 (16.7)
Eczema	2 (16.7)	1 (8.3)
Excoriation	2 (16.7)	0 (0)

N (%); MedDRA/J ver. 15.0

There were no deaths, serious adverse events, or adverse events leading to discontinuation.

disorder, have received leptin replacement therapy for ≥2 months, have experienced improvement from baseline in at least one of diabetes mellitus(symptoms of diabetes mellitus and casual blood glucose level ≥200 mg/dL, fasting blood glucose level ≥126 mg/dL, and 2-hour blood glucose level on an oral glucose tolerance test [OGTT] > 200 mg/dL), hyperinsulinemia (fasting insulin >30 μU/mL), or hypertriglyceridemia (fasting triglyceride > 200 mg/dL), and for which the treatment with metreleptin has been considered to be safe.

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

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

The applicant explained as follows:

No standard treatment has been established in Japan or overseas for glucose and lipid metabolism disorders in patients with lipodystrophy. As symptomatic therapy, lipodystrophy patients have been treated with antidiabetic drugs, antihyperlipidemic drugs, and mecasermin (genetical recombination) approved for treatment of lipoatrophic diabetes mellitus in Japan, but these therapeutic drugs have limited effects.

Leptin, a hormone biosynthesized/secreted by adipose tissue, primarily acts on the hypothalamus to strongly suppress food intake, increase energy expenditure, and inhibit body weight gain.

To elucidate the role of leptin in the pathophysiology of lipodystrophy, the significance of leptin in lipodystrophy was examined using a mouse model of generalized lipodystrophy, a model mouse overexpressing leptin, etc. The results have suggested that the main cause of metabolic disorders in lipodystrophy is lack of leptin (Colombo C, et al. *Diabetes.* 2002;51:2727-33) and that leptin could improve glucose metabolism and insulin sensitivity (Ogawa Y et al. *Diabetes.* 1999;48:1822-9).

Metreleptin is an N-methionyl human leptin analog; the replacement therapy with metreleptin has improved metabolic disorders in patients with lipodystrophy and no clinically significant adverse drug reactions have been reported. Therefore, metreleptin is expected to be a drug with a new mechanism of action to improve glucose metabolism disorders and lipid metabolism disorders in patients with lipodystrophy.

PMDA considers as follows:

Since lipodystrophy is a very rare disorder and complicated with severe metabolic disorders and the effect of symptomatic therapy (mecasermin [genetical recombination] and treatment with antidiabetic drugs and antihyperlipidemic drugs) is limited, metreleptin can provide a new treatment option when it is administered to patients who are given a diagnosis of lipodystrophy under the supervision of a specialist. Lipodystrophy is a very rare disorder and it is difficult to say that the disease is well recognized in clinical practice. Hence, PMDA recommends that the applicant should make efforts to raise the awareness of the disease concept and diagnostic approach for lipodystrophy in cooperation with relevant academic societies in the future.

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

PMDA considers as follows:

Only the results from the Japanese investigator-initiated trial (Study KUTR-003-1) are submitted as the efficacy evaluation data and it is difficult to evaluate the efficacy of the drug product by only using the evaluation data in consideration of the rareness of disorder, seriousness of lipodystrophy, and the number of patients enrolled in the Japanese investigator-initiated trial (Study KUTR-003-1). On the other hand, although data are submitted for reference, the number of patients enrolled in the Japanese clinical research (Study KUTR-003-0) and Japanese investigational study (Study KUTR-003-2) is greater than that in the

Japanese investigator-initiated trial (Study KUTR-003-1), and metreleptin has been evaluated for a longer treatment period. Therefore, there is no choice but to evaluate the efficacy of metreleptin based also on the results of these studies.

Since antidiabetic drugs or antihyperlipidemic drugs could be concomitantly used, discontinued or decreased in the Japanese investigator-initiated trial (Study KUTR-003-1), Japanese clinical research (Study KUTR-003-0), and Japanese investigational study (Study KUTR-003-2), PMDA asked the applicant to explain the effect of concomitant drugs on efficacy evaluation.

The applicant responded as follows:
In the Japanese investigator-initiated trial (Study KUTR-003-1) and Japanese clinical research (Study KUTR-003-0), the number of subjects who received metreleptin in combination with antidiabetic drugs and antihyperlipidemic drugs was 2 (Subject Nos. K01 and K02) and 2 subjects (Subject Nos. 04 and 12), respectively. In the Japanese investigator-initiated trial (Study KUTR-003-1) and Japanese clinical research (Study KUTR-003-0), the number of subjects who received metreleptin in combination with antidiabetic drugs was 1 (Subject No. K03) and 6 subjects (Subject Nos. 01, 02, 03, 05, 08, and 10), respectively. In these subjects, antidiabetic drugs and antihyperlipidemic drugs were discontinued or the doses were reduced by approximately 2 months after the start of metreleptin. Of these subjects, 2 subjects (Subject Nos. 02 and 10) in the Japanese clinical research (Study KUTR-003-0) resumed the treatment with antidiabetic drugs that had been discontinued due to worsening of blood glucose control. One subject (Subject No. 03) in the Japanese clinical research (Study KUTR-003-0) had decreased insulin secretion after entering the Japanese investigational study (Study KUTR-003-2), and addition of liraglutide did not result in an improvement in HbA1c levels. All other subjects, except these 3 subjects, did not experience worsening of HbA1c and triglyceride levels after the discontinuation of antidiabetic drugs and antihyperlipidemic drugs.

PMDA considers as follows:
No control group was used in the Japanese investigator-initiated trial (Study KUTR-003-1), Japanese clinical research (Study KUTR-003-0), and Japanese investigational study (Study KUTR-003-2), and the efficacy of metreleptin was evaluated by comparing the parameters before and after metreleptin treatment in these studies. Ideally, it is desirable to restrict the use of concomitant drugs affecting the efficacy endpoint (especially, antidiabetic drugs and antihyperlipidemic drugs) or changes in their doses during the treatment with metreleptin. However, because the number of Japanese patients with lipodystrophy is very limited, lipodystrophy is expected to be treated concomitantly with antidiabetic drugs and antihyperlipidemic drugs as appropriate rather than metreleptin alone in clinical practice, and no standard treatment has been established, it is unavoidable that the study was conducted in designs mentioned above. Based on the above, PMDA assessed an improvement in glucose and lipid metabolism disorders as shown below.

4.(iii).B.(2).1) Improvement in glucose metabolism disorders

PMDA considers as follows:

HbA1c, the primary efficacy endpoint in the Japanese investigator-initiated trial (Study KUTR-003-1), was found to improve from baseline or be similar to the baseline value (< 5.8%) at Week 20 in all subjects. Of the 4 enrolled subjects, 2 subjects discontinued concomitant antidiabetic drugs, which had been used since before the start of treatment with metreleptin, after starting the metreleptin treatment. The 2 subjects did not experience worsening of HbA1c levels after the discontinuation of concomitant medication [Table 22]. The HbA1c levels over time in each subject in the Japanese clinical research (Study KUTR-003-0) were as shown in Table 31 and HbA1c levels over time of each subject in the Japanese investigational study (Study KUTR-003-2) in Table 32. Although worsening of HbA1c levels was observed in some subjects, the effect lasted for a long time in some patients.

Table 31. HbA1c levels over time in each subject in the Japanese clinical research (Study KUTR-003-0)

Timepoint		Dose[a] (mg/kg)	Subject No. (sex/age)										
			01 (female/ 11 years)[e]	02 (male/ 29 years)[f]	03 (male/ 19 years)[g]	04 (female/ 16 years)[h]	05 (female/ 22 years)[i]	06 (female/ 15 years)	08 (female/ 33 years)[j]	09 ([redacted] years)	10 ([redacted] years)[k]	11 ([redacted] years)	12 ([redacted] years)[l]
Baseline		-	10.0	10.3	8.8	7.9	12.4	6.8	10.2	8.6	9.3	5.1	6.7
Year 1	Month 1	0.01[b] 0.015[b] 0.02[b]	7.3	7.2	6.3	5.9	7.0	7.1	6.0[o]	7.3	6.5	4.8	5.8
	Month 2	0.02[c] 0.03[c] 0.04[c]	5.8	6.6	5.8	5.1	7.1	6.4	-	5.9	5.8	4.7	5.3
	Month 4		4.8	7.1	5.8	5.3	8.9[n]	5.7	-	5.4	7.0	4.7	5.4
	Month 6		6.5	6.4	6.1	4.9	-	6.2	-	5.7	6.1	4.7	6.0
	Month 8		6.3	6.5	7.1	5.1	-	5.9	−	5.3	5.9	4.8	6.1
	Month 12		6.4	6.7	7.4	4.4	-	5.8	−	5.7	5.3	4.9	5.9
Year 2	Month 6		6.8	6.7	8.9	5.9	-	6.1	-	ND	5.8	4.7	5.4[m]
	Month 12		5.8	6.6	10.0	5.3	-	6.1	-	5.4	5.9[p]	4.9	-
Year 3	Month 6		6.3	6.4	9.9	4.7	-	ND	-	5.9	-	5.0[m]	-
	Month 12		5.3	6.4	9.7	4.3	-	5.8	-	5.5	-	-	-
Year 4	Month 6	0.04[d] 0.06[d] 0.08[d]	5.8	7.4	9.7	4.6	-	ND	-	5.6	-	-	-
	Month 12		5.6	10.1	9.8	ND	-	6.5	-	5.4	-	-	-
Year 5	Month 6		5.0	11.2	9.8	4.2	-	6.2	-	5.9	-	-	-
	Month 12		5.5	11.2	9.9	4.5	-	6.6	-	5.6	-	-	-
Year 6	Month 6		5.0	10.4	10.4	4.9	-	6.9	-	5.8[m]	-	-	-
	Month 12		5.4	13.0	10.6	4.9	-	6.8	-	-	-	-	-
Year 7	Month 6		ND	ND	11.8	5.3	-	7.7	-	-	-	-	-
	Month 12		5.4	10.2	11.3	4.8	-	6.4[m]	-	-	-	-	-
Year 8	Month 6		6.2	11.2	10.5	5.1[m]	-	-	-	-	-	-	-
	Month 12		6.2	11.9	ND	-	-	-	-	-	-	-	-
Year 9	Month 6		6.2	12.1[m]	4.7[m]	-	-	-	-	-	-	-	-
	Month 12		5.7[m]	-	-	-	-	-	-	-	-	-	-

Unit, % (JDS value); -, not applicable; ND, not detected (missing data)

a) Upper column, male subjects; middle column, female subjects (<18 years); lower column, female subjects (aged ≥18 years)
b) An estimated dose at which 50% of normal serum leptin concentration is achieved in each subject
c) An estimated dose at which 100% of normal serum leptin concentration is achieved in each subject
d) An estimated dose at which 200% of normal serum leptin concentration is achieved in each subject
e) Pioglitazone hydrochloride (15 mg/day) had been administered concomitantly since before the start of treatment and it was discontinued 13 days after the start of treatment.
f) Voglibose (0.6 mg/day) and glibenclamide (1.25-5.00 mg/day) had been administered concomitantly since before the start of treatment and they were discontinued 50 and 20 days after the start of treatment, respectively. Then, the patient started to receive voglibose (0.4-0.6 mg/day) again 3040 days after the start of treatment and was also given sitagliptin phosphate hydrate (50 mg/day) 3391 days after the start of treatment.
g) Insulin (2-67 units/day) had been administered concomitantly since before the start of treatment and it was discontinued 65 days after the start of treatment. Then, the patient started to receive insulin (2-58 units/day) again 3068 days after the start of treatment and was also given liraglutide (genetical recombination) (0.3-0.6 mg/day) 3117 days after the start of treatment.
h) Pioglitazone hydrochloride (15-30 mg/day) had been administered concomitantly since before the start of treatment and it was discontinued 28 days after the start of treatment.
i) Pioglitazone hydrochloride (15-45 mg/day) had been administered concomitantly since before the start of treatment.
j) Insulin (4-26 units/day) had been administered concomitantly since before the start of treatment and it was discontinued 52 days after the start of treatment.
k) Pioglitazone hydrochloride (30 mg/day) and insulin (6-30 units/day) had been administered concomitantly since before the start of treatment and they were discontinued 13 and 9 days after the start of treatment, respectively. Metformin hydrochloride (750 mg/day) had also been administered concomitantly since before the start of treatment.
l) Pioglitazone hydrochloride (15 mg/kg) and insulin (4 units/day) had been administered concomitantly since before the start of treatment and they were discontinued 53 and 37 days after the start of treatment, respectively. Metformin hydrochloride (250-500 mg/day) had also been administered concomitantly since before the start of treatment.
m) The patients switched to the Japanese investigational study (Study KUTR-003-2).
n) The study was discontinued 129 days after the start of treatment in the opinion of the physician.
o) The study was discontinued 192 days after the start of treatment due to adverse events.
p) The study was discontinued 793 days after the start of treatment due to surgery for bile duct carcinoma.

Table 32. HbA1c levels over time in each subject in the Japanese investigational study (Study KUTR-003-2)

Time point	Subject No. (sex/age/Subject No. in the preceding study)											
	1 ([redacted] years/12)	2 (female/24 years/04)	3 (female/20 years/01)	4 ([redacted] years/11)	5 (male/38 years/02)	6 (female/18 years/K01)	7 ([redacted] years/09)	8 (female/ 24 years/ K02)	9 (female/ 11 years/ K03)	10 (male/ 27 years/ 03)	11 (female/ 22 years/ 06)	12 (male/ 6 years/ K04)
Before switch	5.5	5.1	5.7	5.0	9.2	4.8	5.8	6.4	5.4	4.7	6.4	5.2
Month 2	5.5	4.7	ND	5.0	8.7	5.5	5.7	7.2	5.5	8.7	6.4	4.9
Month 4	5.9	4.4	4.9	5.0	8.1	5.1	5.6	6.4	6.0	9.7	6.8	5.3
Month 6	5.4	4.5	4.8	5.1	8.3	5.4	5.7	6.8	5.8	9.5	7.0	5.1
Month 8	5.4	4.5	4.9	5.1	6.9	5.4	5.8	6.6	5.5	9.4	7.0	5.0[b)]
Month 10	5.4	4.5	4.8	5.2	6.9	5.4	5.9	6.6	5.3	11.1	7.0	4.9
Month 12	5.3	4.8	4.9	4.9	8.7	5.2	5.8	7.4	5.7	9.9[b)]	7.3	-
Month 14	5.5	4.8	4.9[a) b)]	5.1	9.8[b)]	5.1	5.5	7.3	-	-	-	-
Month 16	5.5	4.8	4.9	5.0[b)]	11.7	5.0	5.9	-	-	-	-	-
Month 18	5.3	4.8[b)]	4.9	5.0	-	-	-	-	-	-	-	-
Month 20	5.1[b)]	-	-	-	-	-	-	-	-	-	-	-

Unit, % (JDS value); -, not applicable; ND, not detected (missing data)
See the footnote of Table 31 for concomitant drugs.
a) No measurement was available for an acceptable range (scheduled date of examination calculated from the registration date ± 30 days) and the most recent value was used.
b) The drug product was changed from the drug substance manufactured by [redacted] to that manufactured by [redacted].

PMDA asked the applicant to explain the cause of worsening of HbA1c levels in the subjects (Japanese clinical research [Study KUTR-003-0], Subject No. 02 [Subject No. 5 in the Japanese investigational study, Study KUTR-003-2]; Japanese investigational study [Study KUTR-003-2], Subject No. 10).

The applicant responded as follows:

The Subject No. 02 in the Japanese clinical research (Study KUTR-003-0) did not visit the study center for the subject's personal reasons and administration was missed out multiple times, and the rate of treatment compliance was about 20%. However, after being switched to the Japanese investigational study (Study KUTR-003-2) (Subject No. 5), metreleptin had become self injectable. This brought the treatment compliance rate up to about 100%, followed by an improvement in blood glucose control. However, due to a decrease in the compliance rate, worsening of HbA1c levels was observed at Months 14 and 16 in the Japanese investigational study (Study KUTR-003-2). The Subject No. 10 in the Japanese investigational study (Study KUTR-003-2) discontinued metreleptin and was hospitalized for the treatment of viral fulminant hepatitis before the switch to the Japanese investigational study (Study KUTR-003-2). During the hospitalization, nutritional control had yielded an improvement in blood glucose control, but the subject did not follow dietary recommendations after discharge and blood glucose control exacerbated to the state observed before hospitalization. Then, liraglutide was administered concomitantly because of a decrease in insulin secretion.

PMDA considers as follows:

With respect to blood glucose control in individual patients, the effect lasted for a long time in some subjects, indicating that metreleptin is expected to improve glucose metabolism disorders in lipodystrophy. However, based on the fact that these results were obtained from a very limited number of subjects with lipodystrophy and noncompliance with diet therapy resulted in exacerbation of HbA1c levels in some subjects, it is necessary to include a caution statement on treatment of glucose metabolism disorders (diet therapy, exercise therapy, and combination of existing antidiabetic drugs according to the patient's symptoms) in the package insert and to collect information on an improvement in glucose metabolism disorders via post-marketing surveillance. The above conclusion will be finalized, taking account of comments from the Expert Discussion.

4.(iii).B.(2).2) Improvement in lipid metabolism disorders

PMDA considers as follows:

The triglyceride levels over time, the efficacy endpoint related to lipid metabolism disorders in the Japanese investigator-initiated trial (Study KUTR-003-1), was found to improve from baseline or be similar to the baseline value (< 80 mg/dL) at Week 20 in 3 of 4 subjects. Although worsening of triglycerides was observed in 1 (Subject No. K04) of 4 subjects, it was attributed to concomitant use of corticosteroids (dexamethasone elixir) for 11 days due to exacerbation of sinusitis. Then, after being switched to the Japanese investigational study (Study KUTR-003-2) (Subject No. 12), triglycerides improved. Two out of 4 subjects discontinued concomitant antihyperlipidemic drugs, which had been used since before the start of treatment with metreleptin, after starting the metreleptin treatment, and they did not experience exacerbation of triglycerides after the discontinuation [Table 23]. The triglyceride levels over time in each subject in the Japanese clinical research (Study KUTR-003-0) were as shown in Table 33 and those of triglycerides in each subject in the Japanese investigational study (Study KUTR-003-2) in Table 34. Although exacerbation of triglycerides was observed in some subjects, the effect lasted for a long time in other subjects.

Table 33. Triglyceride levels over time in each subject in the Japanese clinical research (Study KUTR-003-0)

Timepoint		Dose[a] (mg/kg)	Subject No. (sex/age)										
			01 (female/ 11 years)	02 (male/2 9 years)	03 (male/1 9 years)	04 (female/ 16 years)[e]	05 (female/ 22 years)	06 (female/ 15 years)	08 (female/ 33 years)	09 ([redacted] years)	10 ([redacted] years)	11 ([redacted] years)	12 ([redacted] years)[f]
Baseline		-	1941	69	115	1246	254	89	64	227	63	220	61
Year 1	Month 1	0.01[b] 0.015[b] 0.02[b]	307	50	115	596	233	131	99	71	61	87	54
	Month 2	0.02[c] 0.03[c] 0.04[c]	122	51	113	331	164	53	64[i]	51	53	92	67
	Month 4	0.04[d] 0.06[d] 0.08[d]	205	55	86	171	125[h]	68	-	37	80	94	101
	Month 6		180	58	100	223	-	33	-	81	79	73	108
	Month 8		112	73	147	222	-	34	-	25	47	159	85
	Month 12		136	70	55	58	-	31	-	102	136	137	73
Year 2	Month 6		125	42	69	281	-	49	-	ND	195	111	70[g]
	Month 12		260	48	69	183	-	30	-	ND	51[j]	97	-
Year 3	Month 6		112	54	66	96	-	ND	-	ND	-	87[g]	-
	Month 12		162	57	56	115	-	39	-	48	-	-	-
Year 4	Month 6		173	56	81	196	-	ND	-	48	-	-	-
	Month 12		71	44	61	ND	-	39	-	76	-	-	-
Year 5	Month 6		86	68	89	187	-	39	-	202	-	-	-
	Month 12		72	51	80	124	-	61	-	ND	-	-	-
Year 6	Month 6		281	62	85	243	-	54	-	52[g]	-	-	-
	Month 12		101	92	104	349	-	37	-	-	-	-	-
Year 7	Month 6		ND	ND	166	130	-	45	-	-	-	-	-
	Month 12		101	191	207	134	-	44[g]	-	-	-	-	-
Year 8	Month 6		79	82	139	166[g]	-	-	-	-	-	-	-
	Month 12		87	161	ND	-	-	-	-	-	-	-	-
Year 9	Month 6		122	82[g]	158[g]	-	-	-	-	-	-	-	-
	Month 12		106[g]	-	-	-	-	-	-	-	-	-	-

Unit, mg/dL; -, not applicable; ND, not detected (missing data)

a) Upper column, male subjects; middle column, female subjects (aged <18 years); lower column, female subjects (aged ≥18 years)
b) An estimated dose at which 50% of normal serum leptin concentration is achieved in each subject
c) An estimated dose at which 100% of normal serum leptin concentration is achieved in each subject
d) An estimated dose at which 200% of normal serum leptin concentration is achieved in each subject
e) Pravastatin sodium (10-40 mg/day) and bezafibrate (200-400 mg/day) had been administered concomitantly since before the start of treatment and they were discontinued 57 and 120 days after the start of treatment, respectively. Then, the subject started to receive bezafibrate (600 mg/day) again 2337 days after the start of treatment and it was discontinued 2348 days after the start of treatment.
f) Bezafibrate (200-400 mg/day) had been administered concomitantly since before the start of treatment and it was discontinued 67 days after the start of treatment.
g) The subjects switched to the Japanese investigational study (Study KUTR-003-2).
h) The study was discontinued 129 days after the start of treatment in the opinion of the physician.
i) The study was discontinued 192 days after the start of treatment due to adverse events.
j) The study was discontinued 793 days after the start of treatment due to surgery for bile duct carcinoma.

Table 34. Triglyceride levels over time in each subject in the Japanese investigational study (Study KUTR-003-2)

Time point	Subject No. (sex/age/Subject No. in the preceding study)											
	1 ([redacted] years/12)	2 (female/ 24 years/04)	3 (female/ 20 years/01)	4 ([redacted] years/11)	5 (male/38 years/02)	6 (female/18 years/K01)	7 ([redacted] years/09)	8 (female/24 years/K02)	9 (female/11 years/K03)	10 (male/27 years/03)	11 (female/22 years/06)	12 (male/6 years/K04)
Before switch	43	166	106	200	45	62	52	204	77	158	44	382
Month 2	74	220	−	112	91	74	183	181	44	171	50	187
Month 4	79	462	56	90	86	116	174	253	65	118	65	48
Month 6	57	179	49	84	49	436	129	199	52	102	79	70
Month 8	66	188	59	207	42	146	136	123	126	249	84	59[b)]
Month 10	50	259	55	140	63	99	63	181	43	340	57	53
Month 12	65	288	52	99	58	206	82	78	186	159[b)]	49	-
Month 14	48	173	73[a) b)]	116	98[b)]	87	91	219	-	-	-	-
Month 16	45	235	59	164[b)]	113	155	127	-	-	-	-	-
Month 18	58	355[b)]	92	257	-	-	-	-	-	-	-	-
Month 20	39[b)]	-	-	-	-	-	-	-	-	-	-	-

Unit, mg/dL; -, not applicable

See the footnote of Table 33 for concomitant drugs.

a) No measurement was available for an acceptable range (scheduled date of examination calculated from the registration date ± 30 days) and the most recent value was used.

b) After this timepoint, the drug product was changed from the drug substance manufactured by [redacted] to that manufactured by [redacted].

Based on the above, the drug product is expected to improve lipid metabolism disorders in lipodystrophy. However, based on the fact that these results were obtained from a very limited number of subjects with lipodystrophy, it is necessary to include a basic caution statement on treatment of lipid metabolism disorders (diet therapy and exercise therapy) in the package insert and to collect information on an improvement in lipid metabolism disorders via post-marketing surveillance. The above conclusion will be finalized, taking account of comments from the Expert Discussion.

4.(iii).B.(2).3) Effect of antibodies

The applicant explained as follows:

In the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769)[64] and foreign Amylin-sponsored study (Study FHA101)[65], 31 of 35 subjects in whom antibody titer had been determined were tested positive for antibodies

[64] An open-label, uncontrolled study of foreign lipodystrophy patients with glucose metabolism disorders and/or lipid metabolism disorders was conducted. In Study [redacted]1265, male subjects were to receive subcutaneous metreleptin 0.01 mg/kg for the first 1 month, 0.02 mg/kg from Months 1 to 2, and 0.04 mg/kg after Month 3 twice daily in divided doses. Female subjects aged <18 years were to receive subcutaneous metreleptin 0.015 mg/kg for the first 1 month, 0.03 mg/kg from Months 1 to 2, 0.06 mg/kg after Month 3 twice daily in divided doses, and those aged ≥18 years were to receive subcutaneous metreleptin 0.02 mg/kg for the first 1 month, 0.04 mg/kg from Months 1 to 2, and 0.08 mg/kg after Month 3 twice daily in divided doses. The treatment period continued until the switch to Study 20[redacted]0769. In Study 20[redacted]0769, the same dosage regimen as Study [redacted]1265 was used, but after the revision, male subjects or female subjects aged ≤ 9 years were to receive subcutaneous metreleptin 0.015 mg/kg for the first 1 week, 0.03 mg/kg from Weeks 1 to 2, and 0.06 mg/kg after Week 3 once or twice daily in divided doses, and female subjects aged ≥10 years were to receive subcutaneous metreleptin 0.02 mg/kg for the first 1 week, 0.04 mg/kg from Weeks 1 to 2, and 0.08 mg/kg after Week 3 once or twice daily in divided doses. The treatment period was 2 years and after the treatment period, metreleptin treatment could be continued under compassionate use.

[65] An open-label, uncontrolled study of foreign lipodystrophy patients with diabetes mellitus and/or hypertriglyceridemia was conducted. Subjects were to receive subcutaneous metreleptin 0.02 mg/kg for the first 1 month twice daily in divided doses. Then, the dose was increased to 0.04 mg/kg and adjusted according to clinical response. The dosing frequency could also be changed to once daily 1 year after the start of treatment. The treatment period was to last until the discontinuation of the study for safety concerns.

and specific inhibitory activity was weak and there was no reduction in efficacy in all subjects, indicating no production of neutralizing antibodies.

PMDA considers as follows:
The applicant's explanation is acceptable, but it is necessary to examine the effect on antibody expression and efficacy via post-marketing surveillance since there is very limited clinical experience with metreleptin in treating lipodystrophy in Japan and overseas.

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

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

The applicant explained as follows:
Hypoglycemia was reported in 1 of 4 subjects (25.0%) in the Japanese investigator-initiated trial (Study KUTR-003-1), 8 of 55 subjects (14.5%) in the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769), and 1 of 10 subjects (10.0%) in the foreign Amylin-sponsored study (Study FHA101). Hypoglycemia reported in the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769) occurred only in patients receiving insulin therapy, regardless of whether or not they used oral antidiabetic drugs, suggesting that great improvement in insulin sensitivity associated with the treatment with metreleptin. There is a possibility that appropriate reduction of insulin dose was not conducted according to the improvement in insulin sensitivity. Based on the above, careful administration of metreleptin will be recommended in the package insert for patients who are at risk of hypoglycemia, those receiving concomitant drugs that reinforce the glucose lowering effect, and those who are at risk of hypoglycemia-related injury. A caution including information on how to manage hypoglycemia will be included in the package insert.

PMDA considers as follows:
The applicant's explanation is acceptable, but hypoglycemia needs to be monitored carefully not only for concomitant insulin products but also for concomitant use of metreleptin with antidiabetic drugs and it is necessary to appropriately provide cautions in the package insert. A caution should also be exercised when reducing the dose of antidiabetic drugs after the start of treatment with metreleptin. It is necessary to continue to collect information on the occurrence of hypoglycemia and concomitant use of antidiabetic drugs via post-marketing surveillance.

4.(iii).B.(3).2) Acute pancreatitis

The applicant explained as follows:
Acute pancreatitis was reported in 5 of 55 subjects (9.1%) in the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769) and 1 of 10 subjects (10.0%) in the foreign Amylin-sponsored study (Study FHA101). All of the 5 subjects with acute pancreatitis in the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769) had a history of pancreatitis and hypertriglyceridemia. Acute pancreatitis occurring in 4 subjects was reported as a serious adverse event but a causal relationship to metreleptin was ruled out. The events in 3 of the 4 subjects were considered to be due to sudden discontinuation of metreleptin or noncompliance with the treatment regimen and the event in the remaining 1 subject was due to discontinuation of metreleptin with adjustment.

Acute pancreatitis occurring in 1 subject in the foreign Amylin-sponsored study (Study FHA101) was reported as a serious adverse event, but a causal relationship to metreleptin was ruled out. Patients with lipodystrophy are predisposed to serious life-threatening acute pancreatitis due to severe hypertriglyceridemia (Yadav D, et al. *J Clin Gastroenterol.* 2003;36:54-62). Therefore, treatment with metreleptin may reduce risks if a decrease in triglycerides is observed with the treatment, but patients with a history of pancreatitis or those with persistent hypertriglyceridemia are considered to be at increased risk of acute pancreatitis. Thus, the package insert will recommend careful administration in patients with a history of pancreatitis or hypertriglyceridemia since acute pancreatitis may develop after abrupt discontinuation of metreleptin treatment.

PMDA concluded that the applicant's explanation is acceptable.

4.(iii).B.(3).3) T-cell lymphoma

The applicant explained as follows:

T-cell lymphoma was reported in 2 of 55 subjects (3.6%) in the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769). These patients had hematological abnormalities before treatment with metreleptin. Although there is no evidence that indicates treatment with metreleptin increases the risk of onset of T-cell lymphoma, leptin has been reported to, via its receptors, stimulate the production of pro-inflammatory cytokines in cultured monocytes and that of Th1 type cytokines in lymphocytes (Sanchez-Margalet V, et al. *Clin Exp Immunol.* 2003;133:11-9). Based also on the report that leptin inhibits stress-induced apoptosis of T cells and monocytes, it cannot be ruled out that metreleptin may have caused the progression of T-cell lymphoma. Therefore, the package insert will recommend the careful administration of metreleptin in patients with severe hematological disorders.

PMDA considers as follows:

The 2 subjects, in whom T-cell lymphoma was reported in the foreign studies, both had acquired generalized lipodystrophy. One subject had neutropenia of unknown etiology before treatment with metreleptin, and the other subject was diagnosed with chronic idiopathic neutropenia before treatment with metreleptin, accompanied by significant bone marrow hyperplasia with erythroid predominance and a reactive lymph node. Both subjects have the possibility of having lymphoproliferative disorders. Acquired generalized lipodystrophy is often associated with autoimmune disease and these patient factors may have affected the occurrence of T-cell lymphoma. Adverse events related to hematoproliferative disorders including T-cell lymphoma were not observed in the Japanese studies, but leptin is known to have an effect on the immune system. Hence, it is necessary to appropriately provide a caution on those adverse events in the package insert and to continue to collect information via post-marketing surveillance.

4.(iii).B.(3).4) Effect of unphysiologically high serum leptin concentrations

(a) Effect on the diencephalon-hypothalamo-hypophyseal system

Since some subjects had serum leptin concentrations greatly exceeding the normal range in the Japanese investigator-initiated trial (Study KUTR-003-1) and Japanese clinical research (Study KUTR-003-0), PMDA

asked the applicant to explain the possibility that increased leptin levels associated with metreleptin treatment may affect the diencephalon-hypothalamo-hypophyseal system and the growth and development of children.

The applicant responded as follows:
There were no adverse events with the potential impact on hormone secretion in the Japanese investigator-initiated trial (Study KUTR-003-1). In the Japanese clinical research (Study KUTR-003-0), as "Endocrine disorders" in the category of MedDRA system organ class (SOC), 2 episodes of hyperthyroidism were reported in 1 subject (9.1%), 1 episode of goitre and thyroid cyst were observed in 1 subject (9.1%) each, and 2 episodes of dysmenorrhoea occurred in 1 subject (9.1%) as "Reproductive system and breast disorders" in the SOC; all of the events were mild. The serum leptin concentrations measured right around the time of occurrence were in the range of 1.92 to 7.00 ng/mL in the subjects with hyperthyroidism, 15.03 ng/mL in the subjects with goitre or thyroid cyst, and 1.03 to 2.63 ng/mL in the subject with dysmenorrhoea, and they were not markedly high. In the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769), as "Endocrine disorders" in the SOC, 1 episode of autoimmune thyroiditis (mild) occurred in 1 subject (1.8%), 1 subject of euthyroid sick syndrome (mild) in 1 subject (1.8%), and 1 subject of goitre (mild) in 1 subject (1.8%). As "Reproductive system and breast disorders" in the SOC, 2 episodes of amenorrhoea (moderate) occurred in 1 subject (1.8%), 1 episode of cervical cyst (mild) in 1 subject (1.8%), 2 episodes of menstruation irregular (moderate) in 2 subjects (3.6%), 2 episodes of ovarian cyst (mild) in 2 subjects (3.6%), and 3 episodes of ovarian cyst (moderate) in 3 subjects (5.5%). However, the incidence rates of these events were as low as in the range of 1.8% to 5.5%. The serum leptin concentrations measured at the time of occurrence ranged from 0.79 to 62.20 ng/mL and they were not generally high.

As for the hormones secreted by the diencephalon-hypothalamo-hypophyseal system, results from the Japanese and foreign studies have indicated no clear correlation of serum leptin concentrations with adrenocorticotropic hormone (ACTH), thyroid-stimulating hormone (TSH), luteinizing hormone (LH), and follicle-stimulating hormone (FSH). Leptin facilitates secretion of gonadotropin-releasing hormone, which would facilitate the secretion of LH and FSH; however, its effect has not been observed. It has not been reported that subjects with increased leptin levels develop adverse events that may be attributed to abnormal hormone secretion by the diencephalon-hypothalamo-hypophyseal system, including growth hormone (GH) and prolactin (PRL).

However, leptin is responsible for the control of body weight and energy balance, including the regulation of hormone secretion from the hypothalamic-anterior-pituitary-system, and it is essential for the maintenance of reproductive endocrine function (Sakuma Y. *Journal of Japan Society for the Study of Obesity*. 2001;7:93-7); therefore, the possibility that long-term persistence of increased leptin levels may result in early development of secondary sexual characteristics cannot be denied. In addition, GH has a serious effect on not only bone growth but also lipid metabolism, and serum GH secretion has negative correlation to body fat and leptin concentrations in obese children. The direct action of leptin on the peripheral nervous system enhances osteogenesis and in contrast, leptin inhibits osteogenesis through the central nervous system, thereby maintaining osteogenesis through reduced central nervous system action during starvation and increasing

osteogenesis through increased peripheral nervous system action during satiation (Sugihara S. *Bioclinica.* 2003;18:54-8). As described previously, both peripheral and central nervous system actions of leptin are important to the growth of children and the possibility that the persistence of hyperleptinemia may lead to early development of secondary sexual characteristics cannot be ruled out. Even so, leptin is unlikely to abnormally enhance the growth and development of children.

PMDA considers as follows:
The applicant's response is acceptable. However, since leptin regulates reproductive function by facilitating the secretion of gonadotropin-releasing hormone and promotes reproductive function through its central nervous system, the risk of precocious puberty cannot be denied especially in children. Thus, it is necessary to continue to collect information on the effect on the diencephalon-hypothalamo-hypophyseal system via post-marketing surveillance.

(b) Effect on other tissues

Based on the fact that leptin receptors are widely present in the body, PMDA asked the applicant to explain the effect of metreleptin on the cranial nerve system, immune system, and cardiovascular system.

The applicant responded as follows:
Leptin receptors are expressed not only in the hypothalamus but also in kidneys, lungs, adrenal glands, and blood cells, etc. In addition to leptin receptor-mediated appetite suppression and body weight regulation by the satiety center located in the ventromedial hypothalamic nucleus, leptin is considered to be involved in a wide range of effects (Fujita Y. *Clinical Immunology.* 2004;41:711-3). Leptin receptors are also expressed in peripheral immunocompetent cells (Sato [Mito] N. *Japanese Journal of Clinical Medicine.* 2010;68:422-6).

To examine the effect of metreleptin on the cranial nerve system, adverse events falling under the category of "Nervous system disorders" in the SOC were analyzed by event and severity. The adverse events with high incidences were headache (50.0%, 2 of 4 subjects) in the Japanese investigator-initiated trial (Study KUTR-003-1) and headache (63.6%, 7 of 11 subjects) in the Japanese clinical research (Study KUTR-003-0), and no corresponding adverse events were reported in the Japanese investigational study (Study KUTR-003-0). Since the severity of each adverse event was mild and most of the events resolved, clinically evident nervous system disorders have not been reported.

It has been reported that recombinant mouse leptin may induce autoimmunity in pancreatic beta cells in nonobese diabetic (NOD) mice, a model of autoimmune diabetes mellitus (Matarese G, et al. *Diabetes.* 2002;51:1356-61), and compared with the control group, type 1 diabetes mellitus occurs more significantly in the leptin group. However, the potential of autoimmunity to pancreatic beta cells induced by treatment with metreleptin in humans remains unclear.

It has been made clear that blood pressure increases in transgenic mice overexpressing leptin due to sympathicotonia, suggesting the possibility that leptin may cause hypertension as an obesity complication

(Ebihara K. *Japanese Journal of Clinical Medicine.* 2009;67:391-5). In the Japanese studies, only 1 patient (Subject No. 10) in the Japanese clinical research (Study KUTR-003-0) experienced increased blood pressure (mild) as an adverse event; increased blood pressure (mild) occurred on Days 94 and 363 after the start of treatment with metreleptin and the serum leptin concentrations measured at the time closest to the day of occurrence of the event were in the range of 6.88 (Day 59) to 76.83 ng/mL (Day 365). Other subjects had similar serum leptin concentrations, but they did not experience increased blood pressure. Therefore, the risk of increased blood pressure is low even when treatment with metreleptin results in overexpression of leptin.

PMDA considers as follows:
Despite the limitation in available data, results from the safety pharmacology studies of metreleptin have suggested that at least, metreleptin does not have a serious impact on the respiratory system, cardiovascular system, and central nervous system, and the toxicity studies in dogs have indicated that the effect on the immune system is of little relevance to humans. From these results and based on the safety information about the respiratory system, cardiovascular system, and nervous system in the Japanese investigator-initiated trial (Study KUTR-003-1), Japanese clinical research (Study KUTR-003-0), and Japanese investigational study (Study KUTR-003-2), increased leptin levels are not considered to have a serious impact on safety. However, it is necessary to collect information about the effect on other tissues via post-marketing surveillance.

4.(iii).B.(3).5) Effect of antibodies

The applicant explained as follows:
Of 20 subjects who were tested negative for antibodies at baseline in the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769), 12 subjects were tested positive for antibodies and 8 subjects were tested negative after treatment with metreleptin at the time of data cut-off in [redacted] 20[redacted]. The effect on safety was evaluated in these subjects and the occurrence of adverse events with or without antibody expression was as shown in Table 35. Although there are differences in the incidence of adverse events, the number of subjects is low and adverse events specific to antibody expression do not tend to occur.

Table 35. Adverse events with or without antibody expression (at least 2subjects in either of the treatment groups)

Name of the event	Antibody-positive (n = 12)	Antibody-negative (n = 8)
Total	12 (100.0)	8 (100.0)
Abdominal pain	4 (33.3)	0 (0.0)
Ovarian cyst	4 (33.3)	0 (0.0)
Hypoglycaemia	3 (25.0)	2 (25.0)
Dizziness	3 (25.0)	1 (12.5)
Headache	3 (25.0)	1 (12.5)
Alopecia	3 (25.0)	0 (0.0)
Anaemia	2 (16.7)	0 (0.0)
Iron deficiency anaemia	2 (16.7)	0 (0.0)
Nausea	2 (16.7)	1 (12.5)
Pancreatitis	2 (16.7)	0 (0.0)
Vomiting	2 (16.7)	0 (0.0)
Oedema	2 (16.7)	1 (12.5)
Upper respiratory tract infection	2 (16.7)	2 (25.0)
Ear infection	2 (16.7)	1 (12.5)
Vitamin D deficiency	2 (16.7)	0 (0.0)
Muscle spasms	2 (16.7)	0 (0.0)
Paraesthesia	2 (16.7)	1 (12.5)
Proteinuria	2 (16.7)	1 (12.5)
Renal cyst	2 (16.7)	1 (12.5)
Dysuria	2 (16.7)	0 (0.0)
Menstruation irregular	2 (16.7)	0 (0.0)
Fatigue	1 (8.3)	3 (37.5)
Pain in extremity	1 (8.3)	2 (25.0)
Cough	0 (0.0)	2 (25.0)

N (%); MedDRA ver.13.0

PMDA considers as follows:

Although the number of subjects studied was limited, no great difference was observed for safety regardless of antibody expression. However, it is necessary to examine the effect of antibody expression on safety via post-marketing surveillance since there is very limited clinical experience with metreleptin in treating lipodystrophy in Japan and overseas.

4.(iii).B.(4) Indications

Based on the clinical positioning of metreleptin, PMDA asked the applicant to explain the appropriateness of indications.

The applicant responded as follows:

In general, children are less likely to develop diabetes mellitus and control of hypertriglyceridemia can be achieved by strict diet therapy in some cases. However, since patients with lipodystrophy have defects or dysfunction in adipocytes or partial loss of adipocytes, they often have hepatic steatosis caused by the accumulation of fat in the liver even in the cases where symptoms of diabetes mellitus or hypertriglyceridemia are stable. Since insulin resistance is observed before development of diabetes mellitus, early initiation of leptin replacement therapy is expected to prevent fat accumulation in the liver and thus improve prognosis. On the other hand, the number of Japanese patients with partial lipodystrophy is as small as 2 (1 subject in the Japanese investigator-initiated trial [Study KUTR-003-1] [Subject No. K02, female, 23 years, acquired partial lipodystrophy] and 1 subject in the Japanese clinical research [Study KUTR-003-0] [Subject No. 12, [redacted], [redacted] years, familial partial lipodystrophy]). Both subjects had poorly-controlled diabetes mellitus, hypertriglyceridemia, and hepatic steatosis, but leptin replacement therapy enabled the discontinuation of insulin treatment in the subjects. In partial lipodystrophy, partial atrophy of fat may result

in delayed diagnosis until metabolic disorders exacerbates, but it is considered important to start treatment as early as possible. Based on the above, the applicant considers it is appropriate to start metreleptin treatment in patients with insulin-resistant generalized or partial lipodystrophy at an early stage.

PMDA considers as follows:
The Japanese investigator-initiated trial (Study KUTR-003-1) was conducted in lipodystrophy patients with diabetes mellitus or hyperinsulinemia and the Japanese clinical research (Study KUTR-003-0) and Japanese investigational study (Study KUTR-003-2) included lipodystrophy patients with diabetes mellitus, hyperinsulinemia, or hypertriglyceridemia, and subjects with both hyperglycemia and hypertriglyceridemia were not necessarily enrolled in these studies. Even when patients do not develop diabetes mellitus or hypertriglyceridemia, it is possible that they have experienced lack of leptin caused by defects or loss of adipose tissue, which is the main cause of metabolic disorders associated with lipodystrophy. Taking account of the above and the possibility that early initiation of leptin replacement may improve prognosis, it is appropriate to specify "lipodystrophy" as an indication and include the target disease (glucose metabolism disorders, lipid metabolism disorders, etc.) in the "Precautions for Indications" section of the package insert. The appropriateness of the indication and of the inclusion of the target disease in the "Precautions for Indications" will be finalized, taking account of comments from the Expert Discussion.

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

4.(iii).B.(5).1) Dosage regimen

The applicant explained as follows:
Diurnal variations in endogenous leptin are not greater than those in growth hormone and adrenocortical hormone and the diurnal variations even at night time, at which variations are largest, are only approximately 1.5 times those in the daytime. Therefore, in the foreign NIH clinical study (Study [redacted]1265), a twice-daily regimen was selected in consideration of serum metreleptin half-life. However, there are many pediatric patients with lipodystrophy for whom metreleptin is indicated and therefore the efficacy of a once-daily regimen was evaluated with the focus on the reduction in the burden of injections and the rate of continued treatment (Ebihara K, *et.al*. *J Clin Endocrinol Metab.* 2007;92:532-41). As a result, no difference in efficacy was found between the once-daily regimen and twice-daily regimen and the former contributed to the improvement of convenience for patients. The dosage regimen was changed to enable the once-daily regimen to be used in the Japanese clinical research (Study KUTR-003-0).

PMDA considers as follows:
Since (a) there has been no specific problem with the Japanese investigator-initiated trial (Study KUTR-003-1) and Japanese investigational study (Study KUTR-003-2) using the once-daily regimen and (b) the switch from the twice-daily to the once-daily regimen in the Japanese clinical research (Study KUTR-003-0) did not cause major problems, the use of the once-daily regimen is acceptable. The above conclusion will be finalized, taking account of comments from the Expert Discussion.

4.(iii).B.(5).2) Dose

The applicant explained as follows:
In both the Japanese investigator-initiated trial (Study KUTR-003-1) and Japanese clinical research (Study KUTR-003-0), the doses were selected based on the results of the preceding foreign NIH clinical study (Study [redacted]1265). The details of the method for estimating the doses are not clear but its appropriateness was discussed as follows:

To maintain normal serum leptin concentrations in patients with lipodystrophy, they are considered to be required to receive the dose corresponding to the daily production of endogenous leptin in healthy adults. From the reason that the daily production is speculated to be equivalent to the daily elimination of endogenous leptin under normal conditions, the formula (daily production of endogenous leptin = leptin clearance [CL] × area under the daily endogenous leptin concentration-time curve [AUC]) is considered to be established and the following relationship between endogenous leptin concentration and amount of fat has been reported: $\log_{10}$[mean endogenous leptin concentration (ng/mL)] = 0.34 + 0.028 × amount of fat (kg) in men and $\log_{10}$[mean endogenous leptin concentration (ng/mL)] = 0.75 + 0.025 × amount of fat (kg) in women (Saad MF, et al. *J Clin Endocrinol Metab.* 1997;82:579-84). In the foreign phase I subcutaneous study (Study LEPT-[redacted]0272), the daily production of leptin calculated based on leptin's CL/F (assuming a subcutaneous bioavailability [F] of 1) by dose range from 0.013 to 0.020 mg/kg in male subjects and 0.033 to 0.052 mg/kg in female subjects. These values are similar to estimated doses, at which 100% of normal serum leptin concentrations is achieved, in adult male and female subjects (0.02 and 0.04 mg/kg, respectively). Hence, the doses are considered to be estimated appropriately. However, no subjects attained normal serum leptin concentrations at 50% dose in the Japanese studies, not more than half of the number of the subjects achieved normal serum leptin concentrations at 100% dose, and all subjects (4 of 4 subjects) in the Japanese investigator-initiated trial (Study KUTR-003-1) and 9 of 10 subjects in the Japanese clinical research (Study KUTR-003-0) attained normal serum leptin concentrations at 200% dose, indicating the necessity of using 200% dose as a usual dose. For safety, the incidence rates of adverse events reported within <4 months after the start of treatment (including the treatment periods for 50% dose and 100% dose) and from ≥4 months to <1 year (the treatment period for 200% dose) were 90.9% (10 of 11 subjects) and 81.8% (9 of 11 subjects) in the Japanese clinical research (Study KUTR-003-0), and those of adverse drug reactions were 45.5% (5 of 11 subjects) and 18.2% (2 of 11 subjects). The incidence of adverse drug reactions tended to be high within <4 months after the start of treatment, but the trend appeared to be due to injection site reactions reported frequently at the start of subcutaneous treatment. Therefore, the applicant concluded that there was no association between the dose and safety. Based on the above, the usual dose was determined to be 200% dose. Treatment was started with 50% dose in the clinical studies. However, since (a) there is no association between dose and safety and (b) concomitant use of antidiabetic drugs such as insulin may cause hypoglycemia at the start of treatment, treatment will be started with 100% dose, half of the usual dose.

As adipocytes increase in number in females in the development of secondary sexual characteristics, endogenous leptin also increases, thus promoting reproductive function. Females generally start to develop secondary sexual characteristics by the age of 16 years, but reproductive function may not be stable until the

age of around 18 years. Based on the above findings and results of the Japanese studies, the applicant considers that it is appropriate to increase dose in female subjects aged ≥18 years.

PMDA considers as follows:

The applicant explained that the doses had been determined in the Japanese studies by reference to those used in the preceding foreign NIH clinical study (Study [redacted]1265/20[redacted]0769). However, the dose selection for the Japanese studies cannot be justified because (a) the rationale for the dose selection in the NIH clinical study (Study [redacted]1265/20[redacted]0769) is unknown; (b) the doses were changed during the NIH clinical study and the study is being conducted with doses different from those used in the Japanese studies; (c) different doses were not set by age and sex in the foreign Amylin-sponsored study (Study FHA101) started after the NIH clinical study; (d) the fixed doses proposed in the US are 0.06 mg/kg in subjects with a body weight of ≤40 kg and 2.5 mg and 5.0 mg in males and females, respectively, with a body weight of >40 kg; and (e) 100% dose has been determined based on serum leptin concentrations observed at body fat percentage of 20% (males) or 30% (females), which is the upper limit of the normal range for body fat percentage,[62] and it differs from the mean of endogenous serum leptin concentrations in Japanese healthy adult subjects[57] (3.9 ng/mL for male subjects and 7.3 ng/mL for female subjects). The applicant discussed the justification of the dose by the attainment of normal serum leptin concentrations in terms of leptin replacement therapy. However, there are considerable individual differences in endogenous serum leptin concentrations and diurnal variations are known to occur. Therefore, the justification for taking the target serum leptin concentration as the normal level is not evident. In the Japanese studies, administration of 200% dose results in higher serum leptin concentrations in some patients compared with the normal serum leptin concentrations. The effect of increased leptin levels on long-term safety is unknown and it is appropriate to administer the minimum dose required to maintain serum leptin concentrations at the normal level. Based on the above, it is difficult to conclude that the proposed dose is optimal for treatment of patients with lipodystrophy. However, it is unavoidable to use 200% dose as a usual dose since (a) the possibility that serum leptin concentrations measured in the Japanese studies etc. was affected by antibodies, etc. cannot be ruled out, (b) no major safety issues have been identified for Japanese patients with high serum leptin concentrations, (c) it is considered difficult to conduct a dose-finding study of Japanese patients due to a very limited number of patients with lipodystrophy, and (d) the Japanese studies have indicated that metreleptin can be effective for treating lipodystrophy and found no major safety concerns. Regarding the initial dose, although it was studied in healthy adults, the subjects received >200% dose without dose escalation in the foreign phase I studies (Studies LEPT-[redacted]0121 and LEPT-[redacted]0272). No problems were identified in terms of tolerability. Thus, given that appropriate caution is provided for hypoglycemia caused by concomitant antidiabetic drugs at the start of treatment, starting treatment with 100% dose is speculated to cause no major problems. Although the numbers were small, female patients aged <18 years were enrolled in the Japanese studies and the efficacy and safety were evaluated with the proposed dose. Therefore, despite the inadequate rationale for dose selection, it is also considered unavoidable to set the dose according to age. In addition to the appropriateness of the dose, the monitoring method during treatment with metreleptin and the appropriateness of Dosage and Administration section of the package insert will be finalized, taking account of comments from the Expert Discussion.

4.(iii).B.(6) Special populations

4.(iii).B.(6).1) Pediatric use

PMDA considers as follows:

There is no clinical experience with metreleptin in children aged <6 years in Japan, nor metreleptin was also administered to children aged <7 years in the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769) and foreign Amylin-sponsored study (Study FHA101). Based on these facts, it should be cautioned in the package insert that there is no clinical experience with metreleptin in low-birth-weight infants, neonates, nursing infants, or infants aged <6 years and its safety has not been established.

4.(iii).B.(6).2) Use in pregnant, parturient, and lactating women

The applicant explained as follows:

The reproductive and developmental toxicity studies in mice have reported decreases in litter size, low pup survival rates, weight loss, and growth retardation. Therefore, metreleptin should be used in pregnant or potentially pregnant women only if the expected therapeutic benefits outweigh the possible risks associated with the treatment. The excretion of metreleptin in milk is unknown, but because endogenous leptin is detected in milk and the safety of metreleptin in infants has not been established, nursing mothers should discontinue breast-feeding while receiving metreleptin.

PMDA considers as follows:

There is no problem with the applicant's view that metreleptin should be used in pregnant or potentially pregnant women only if the expected therapeutic benefits outweigh the possible risks associated with treatment since lipodystrophy may be associated with serious metabolic disorders. PMDA also finds no problem with the caution for nursing mothers.

4.(iii).B.(6).3) Use in patients with renal impairment and the elderly

PMDA considers as follows:

Since the clinical studies of metreleptin did not include patients with renal impairment, the effect of renal impairment on pharmacokinetics is unknown. On the other hand, results from the non-clinical pharmacokinetic studies have indicated that metreleptin is eliminated primarily by renal excretion in humans through glomerular filtration, and serum leptin concentrations may increase in patients with renal impairment. Therefore, it should be cautioned in the package insert that serum leptin concentrations may increase in patients with renal impairment.

As for use in the elderly, the applicant explained as follows:

Patients aged ≥65 years were not enrolled in the Japanese studies. In contrast, the foreign NIH clinical study (Study [redacted]1265/20[redacted]0769) and foreign Amylin-sponsored study (Study FHA101) included 1 lipodystrophy patient aged ≥65 years each (68 and 67 years at the time of registration, respectively), but there have been no reports of impaired physiological function of endogenous leptin. Thus, although whether or not the physiological function of endogenous leptin is compromised in elderly patients with lipodystrophy remains

unknown, it should be noted that they are apt to develop adverse drug reactions since in general, physiological function declines in the elderly. Therefore, careful administration of metreleptin was recommended in the package insert for elderly patients.

PMDA considers that there is no problem with the caution for use in the elderly.

4.(iii).B.(7) Post-marketing surveillance

The applicant explained as follows:

Since there is limited clinical experience with metreleptin in Japan, post-marketing surveillance (2 years for registration, 2 years for observation, and 40 patients to be followed up) of all patients who received the drug will be conducted until data from a certain number of patients are accumulated. In this post-marketing surveillance, the safety and efficacy of metreleptin will be evaluated in routine medical practice and the effect of long-term treatment on safety will also be examined.

PMDA considers as follows:

Based on the fact that the number of patients with lipodystrophy is very limited, there is limited clinical experience with metreleptin in Japan and overseas, and the effect of prolonged exposure to unphysiologically high leptin concentrations associated with leptin replacement therapy on safety is unknown, registration and observation should take place for a longer period. The details of post-marketing surveillance, including the above conclusion, will be finalized, taking 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 were 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 was conducted in accordance with the provisions of the Pharmaceutical Affairs Act for the data submitted in the new drug application (5.3.5.2-1). Protocol deviations (noncompliance with the requirements for the test schedules) were found at trial sites. Although this finding requiring improvement was noted, applicable subjects were handled properly; therefore, PMDA concluded that the clinical trial is conducted on the whole in compliance with GCP and that 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 efficacy of metreleptin in the treatment of patients with lipodystrophy has been demonstrated and its safety is acceptable in view of its observed benefits. Metreleptin is considered to serve as a new treatment option for lipodystrophy and be of clinical significance. The safety and efficacy of metreleptin need to be investigated via post-marketing surveillance involving all treated patients since the safety and efficacy of metreleptin cannot be fully evaluated due to a very limited number of patients treated in Japan and metreleptin is intended for long-term use.

PMDA considers that metreleptin 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 15, 2013

I. Product Submitted for Registration

[Brand name]	Metreleptin for Subcutaneous Injection 11.25 mg "Shionogi" (changed from Metreleptin for Subcutaneous Injection 11.3 mg "Shionogi")
[Non-proprietary name]	Metreleptin (Genetical Recombination)
[Name of applicant]	Shionogi & Co., Ltd.
[Date of application]	July 27, 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) Efficacy

1) Improvement in glucose metabolism disorders

PMDA considers as follows:

From the results of the HbA1c levels over time in the Japanese investigator-initiated trial (Study KUTR-003-1), Japanese clinical research (Study KUTR-003-0), and Japanese investigational study (Study KUTR-003-2), metreleptin is expected to improve glucose metabolism disorders associated with lipodystrophy. However, based on the fact that these results were obtained from a very limited number of subjects with lipodystrophy and noncompliance with diet therapy resulted in worsening of HbA1c levels in some subjects, it is necessary to include a caution statement on treatment of glucose metabolism disorders (diet therapy, exercise therapy, and combination of existing antidiabetic drugs according to the patient's symptoms) in the package insert and to collect information on an improvement in glucose metabolism disorders via post-marketing surveillance.

The above conclusion of PMDA was supported by the expert advisors. Based on the above, PMDA asked the applicant to consider the inclusion of a caution statement on treatment of glucose metabolism disorders in the package insert, and confirmed that appropriate action was taken by the applicant [for post-marketing surveillance, see "(5) Post-marketing surveillance"].

2) Improvement in lipid metabolism disorders

PMDA considers as follows:

From the results of the triglyceride levels over time in the Japanese investigator-initiated trial (Study KUTR-003-1), Japanese clinical research (Study KUTR-003-0), and Japanese investigational study (Study KUTR-003-2), metreleptin is expected to improve lipid metabolism disorders associated with lipodystrophy. However, based on the fact that these results were obtained from a very limited number of subjects with lipodystrophy, it is necessary to include a basic caution statement on treatment of lipid metabolism disorders (diet therapy and exercise therapy) in the package insert and to collect information on an improvement in lipid metabolism disorders via post-marketing surveillance.

The above conclusion of PMDA was supported by the expert advisors. Based on the above, PMDA asked the applicant to consider the inclusion of a basic caution statement on treatment of lipid metabolism disorders in the package insert, and confirmed that appropriate action was taken by the applicant [for post-marketing surveillance, see "(5) Post-marketing surveillance"].

(2) Safety

1) Hypoglycemia

PMDA considers as follows:

Hypoglycemia needs to be monitored carefully not only for concomitant insulin products but also for concomitant use of metreleptin with antidiabetic drugs and it is necessary to appropriately provide cautions in the package insert. A caution should also be provided on dose reduction of antidiabetic drugs after the start of treatment with metreleptin. It is necessary to continue to collect information on the occurrence of hypoglycemia and concomitant use of antidiabetic drugs via post-marketing surveillance.

The above conclusion of PMDA was supported by the expert advisors. Based on the above, PMDA instructed the applicant to consider the inclusion of caution statements on the occurrence of hypoglycemia and dose reduction of antidiabetic drugs after the start of treatment with metreleptin in the package insert, and confirmed that appropriate action was taken by the applicant [for post-marketing surveillance, see "(5) Post-marketing surveillance"].

2) T-cell lymphoma

PMDA considers as follows:

Two subjects reported in the foreign studies had acquired generalized lipodystrophy. One patient had neutropenia of unknown etiology before treatment with metreleptin and the other patient was diagnosed with chronic idiopathic neutropenia before treatment with metreleptin and significant bone marrow hyperplasia with erythroid predominance and a reactive lymph node were observed. Both patients had the possibility of having lymphoproliferative disorders. In addition, acquired generalized lipodystrophy is frequently complicated with autoimmune disease. These patient factors may have affected the occurrence of T-cell lymphoma. Adverse events related to hematoproliferative disorders including T-cell lymphoma were not observed in the Japanese studies, but since leptin is known to have an effect on the immune system, it is necessary to appropriately provide a caution in the package insert and continue to collect information on the occurrence of T-cell lymphoma via post-marketing surveillance.

The above conclusion of PMDA was supported by the expert advisors. Based on the above, PMDA instructed the applicant to consider the inclusion of a caution on the occurrence of T-cell lymphoma in the package insert, and confirmed that appropriate action was taken by the applicant [for post-marketing surveillance, see "(5) Post-marketing surveillance"].

3) Effect of unphysiologically high serum leptin concentrations

PMDA considers as follows:

Since leptin regulates reproductive function by facilitating the secretion of gonadotropin-releasing hormone and promotes reproductive function through the central nervous system, the risk of precocious puberty cannot be denied especially in children. Thus, it is necessary to continue to collect information on the effect on the diencephalon-hypothalamo-hypophyseal system via post-marketing surveillance. As for the effect on other tissues, though the data are limited to those obtained in specific body parts, results from the safety pharmacology studies of metreleptin have suggested that metreleptin does not have, at least, a serious impact on the respiratory system, cardiovascular system, and central nervous system, and the toxicity studies in dogs have indicated that the effect on the immune system is little relevance to humans. From these results and based on the safety information about the respiratory system, cardiovascular system, and nervous system in the Japanese investigator-initiated trial (Study KUTR-003-1), Japanese clinical research (Study KUTR-003-0), and Japanese investigational study (Study KUTR-003-2), increased leptin levels are not considered to have a serious impact on safety. However, it is necessary to collect information on the effect on other tissues via post-marketing surveillance.

The above conclusion of PMDA was supported by the expert advisors. The expert advisors commented that in the event of adverse events, serum leptin concentrations should be determined under the post-marketing surveillance so that their relationship can be investigated [for post-marketing surveillance, see "(5) Post-marketing surveillance"].

4) Effect of antibodies

PMDA considers as follows:

No great difference was observed for safety regardless of antibody expression. However, it is necessary to examine the effect on antibody expression and safety via post-marketing surveillance since there is very limited clinical experience with metreleptin in treating patients with lipodystrophy in Japan and overseas. The above conclusion of PMDA was supported by the expert advisors [for post-marketing surveillance, see "(5) Post-marketing surveillance"].

5) Safety in the ongoing studies, etc.

As for the ongoing Japanese investigational study (Study KUTR-003-2) and the currently available overseas safety information, PMDA asked the applicant if there were any changes made after the preparation of Review Report (1).

The applicant responded as follows:
No serious adverse events have been reported in the Japanese investigational study (Study KUTR-003-2). In foreign countries, 6 serious adverse drug reactions (hypoglycaemia [2], hypoglycaemic seizure [1], neutralising antibodies [1], pancreatitis [1], and lymphoma [1]) were reported and all adverse drug reactions, except neutralising antibodies and lymphoma, resolved or improved. Based on the above, the applicant considers that no great change has been made to the available information about the safety of metreleptin and no safety concerns have been identified.

PMDA evaluated the available information about the safety of the drug product and confirmed that no changes have been made to the safety assessment of the drug product.

(3) Indications
PMDA considers as follows:
The Japanese investigator-initiated trial (Study KUTR-003-1) was conducted in lipodystrophy patients with diabetes mellitus or hyperinsulinemia and the Japanese clinical research (Study KUTR-003-0) and Japanese investigational study (Study KUTR-003-2) included lipodystrophy patients with diabetes mellitus, hyperinsulinemia, or hypertriglyceridemia, and subjects with both hyperglycemia and hypertriglyceridemia were not necessarily enrolled in these studies. Even when patients did not develop diabetes mellitus or hypertriglyceridemia, it is possible that they have experienced lack of leptin caused by defects or loss of adipose tissue, which is the main cause of metabolic disorders associated with lipodystrophy. Taking account of the above and the possibility that early initiation of leptin replacement may improve prognosis, it is considered appropriate to set “lipodystrophy” as the indication and include the target disease (glucose metabolism disorders, lipid metabolism disorders, etc.) in the “Precautions for Indications” section of the package insert. The above conclusion of PMDA was supported by the expert advisors.

Based on the above, PMDA instructed the applicant to change the indications to “lipodystrophy” and review the Precautions for Indications.

The applicant responded that the indications were to be changed to “lipodystrophy” and the target disease (glucose metabolism disorders, lipid metabolism disorders, etc.) was to be included in the Precautions for Indications section of the package insert to provide caution.

PMDA confirmed that there is no problem with the applicant’s response.

(4) Dosage and administration
1) Dosage regimen
Since there was no specific problem with the Japanese investigator-initiated trial (Study KUTR-003-1) and Japanese investigational study (Study KUTR-003-2) using the once-daily regimen, nor did the switch from the twice-daily to the once-daily regimen in the Japanese clinical research (Study KUTR-003-0) cause major

problems, PMDA considers that the once-daily regimen is acceptable. The above conclusion of PMDA was supported by the expert advisors.

2) Dose

PMDA considers as follows:

With respect to the proposed dose in the US, 100% dose has been determined based on serum leptin concentrations observed at body fat percentage of 20% (males) or 30% (females), which is the upper limit of the normal range. On the other hand, endogenous serum leptin concentrations in foreign healthy adult subjects differ from the mean of endogenous serum leptin concentrations in Japanese healthy adult subjects (3.9 ng/mL for male subjects and 7.3 ng/mL for female subjects), and therefore, dose selection for the Japanese studies cannot be justified. The applicant discussed the justification of dose by the attainment of normal serum leptin concentrations in terms of leptin replacement therapy. However, there are considerable individual differences in endogenous serum leptin concentrations and diurnal variations are known to occur. Therefore, the appropriateness for taking the target serum leptin concentration as the normal level is not evident. In the Japanese studies, administration of 200% dose resulted in higher serum leptin concentrations in some patients compared with normal serum leptin concentrations. The effect of increased leptin levels on long-term safety is unknown and it is appropriate to administer the minimum dose required to maintain serum leptin concentrations at the normal level. Based on the above, it is difficult to conclude that the proposed dose is optimal for treatment of patients with lipodystrophy. However, it is inevitable to use 200% dose as the usual dose since (a) the possibility that serum leptin concentrations measured in the Japanese studies etc. was affected by antibodies, etc. cannot be ruled out, (b) no major safety issues have been identified for Japanese patients with high serum leptin concentrations, (c) it is difficult to conduct a dose-finding study in Japanese patients due to a very limited number of patients with lipodystrophy, and (d) the Japanese studies have indicated that metreleptin can be effective for treating lipodystrophy and found no major safety concerns. Regarding the initial dose, although evaluation was done on healthy adults, subjects received >200% dose without dose escalation in the foreign phase I studies (Studies LEPT-[redacted]0121 and LEPT-[redacted]0272) and no problems were identified with tolerability. Thus, given that appropriate caution is provided to hypoglycemia caused by concomitant antidiabetic drugs at the start of treatment, starting treatment with 100% dose is speculated to cause no major problems. Although the numbers were small, female patients aged <18 years were enrolled in the Japanese studies and the efficacy and safety were evaluated with the proposed dose. Therefore, despite the inadequate rationale for dose selection, it is also inevitable to set the dose according to age.

The above conclusion of PMDA was supported by the expert advisors. The following comments were raised from some of the expert advisors: It is appropriate to reduce dose by the assessment of a physician so that the minimum effective dose can be administered depending on laboratory values such as blood glucose level and triglycerides; and the measurement of serum leptin concentrations is not essential because serum leptin concentrations are not proportionate to therapeutic effect.

Based on the above, PMDA instructed the applicant to consider the inclusion of a caution statement on the occurrence of hypoglycemia, and confirmed that appropriate action was taken by the applicant. PMDA considers that the post-marketing measurement of serum leptin concentrations can be performed, if necessary, at the discretion of a physician and the information about the results may be collected [for post-marketing surveillance, see "(5) Post-marketing surveillance"].

(5) Post-marketing surveillance

PMDA considers that based on the fact that the number of patients with lipodystrophy is very limited and there is limited clinical experience with metreleptin in Japan and overseas, and the effect of prolonged exposure to unphysiologically high leptin concentrations associated with leptin replacement therapy on safety is unknown, registration and observation should take place for a longer period in surveys of all patients who received metreleptin during the reexamination period (10 years scheduled). The above conclusion of PMDA was supported by the expert advisors.

Based on the above, PMDA instructed the applicant to review a plan for post-marketing surveillance.

The applicant responded as follows:

Post-marketing surveillance (8 years for registration and 9 years for observation) of all patients who received metreleptin will be conducted to evaluate the occurrence of adverse drug reactions (including unexpected adverse drug reactions) after the actual use of metreleptin in clinical practice, and potential factors affecting safety, efficacy, etc. In addition to patient demographics (clinical form of lipodystrophy, time of diagnosis, presence or absence of insulin resistance, complications, etc.), prior treatment and concomitant therapy, blood pressure, ECG, laboratory tests (including blood glucose level, HbA1c, and blood triglycerides), hypoglycemia and acute pancreatitis will be priority investigation items in this surveillance for information collection. The information about T-cell lymphoma and adverse events related to the diencephalon-hypothalamo-hypophyseal system, cranial nerve system, immune system, and cardiovascular system will also be collected. The information on serum leptin concentrations and antibody titer will be collected to examine their effects on safety and efficacy.

PMDA accepted the response.

(6) Brand name

To provide accurate information regarding the strength of active ingredient in the drug product, the applicant proposed the following modification to the brand name of the product:

At the time of filing of the product application	After modification (the underlined part was changed)
Metreleptin for Subcutaneous Injection 11.3 mg "Shionogi"	Metreleptin for Subcutaneous Injection 11.<u>25</u> mg "Shionogi"

III. Overall Evaluation

As a result of the above review, PMDA concludes that the product may be approved for the following indication and dosage and administration, along with the following conditions. The re-examination period is 10 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] Lipodystrophy

[Dosage and administration]

The usual dosage is 0.04 mg/kg as metreleptin for male patients, 0.06 mg/kg for female patients aged <18 years, and 0.08 mg/kg for female patients aged ≥18 years, administered subcutaneously once daily.

Treatment should be initiated at a dose of 0.02 mg/kg, 0.03 mg/kg, and 0.04 mg/kg, respectively, and the dose should be increased up to the dose level specified above within about a month.

The dose may be adjusted according to the patient's symptoms.

[Conditions for approval]

The applicant is required to conduct a drug use-results survey involving all patients treated with the product after the market launch until data from a certain number of patients have been accumulated in order to grasp the demographic information of the treated patients, since the product has been studied in only a limited number of patients in the Japanese clinical studies. At the same time, data on the safety and efficacy of the product should be collected without delay and necessary measures should be taken to ensure proper use of the product.