

Report on the Deliberation Results

February 7, 2014

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

[Brand name]	Forxiga 5 mg tablets, Forxiga 10 mg tablets
[Non-proprietary name]	Dapagliflozin Propylene Glycolate Hydrate (JAN*)
[Applicant]	Bristol-Myers K.K.
[Date of application]	March 15, 2013

[Results of deliberation]

In the meeting held on January 24, 2014, the First Committee on New Drugs concluded that the product may be approved and that this result should be reported to the Pharmaceutical Affairs Department of the Pharmaceutical Affairs and Food Sanitation Council.

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

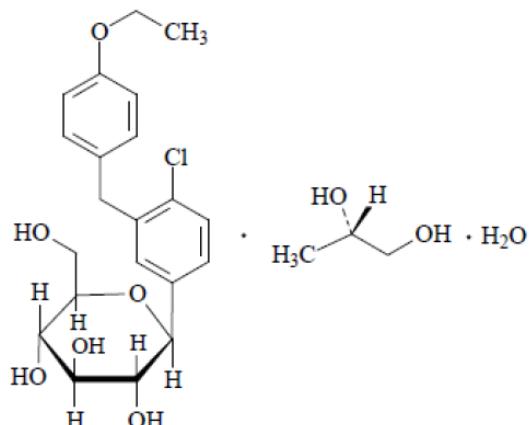
**Japanese Accepted Name (modified INN)*

Review Report

January 6, 2014
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]	Forxiga 5 mg tablets, Forxiga 10 mg tablets
[Non-proprietary name]	Dapagliflozin Propylene Glycolate Hydrate
[Name of applicant]	Bristol-Myers K.K.
[Date of application]	March 15, 2013
[Dosage form/Strength]	Each tablet contains Dapagliflozin Propylene Glycolate Hydrate equivalent to 5 or 10 mg of dapagliflozin.
[Application classification]	Prescription drug, (1) Drug with a new active ingredient
[Chemical structure]	



Molecular formula:	C ₂₁ H ₂₅ ClO ₆ ·C ₃ H ₈ O ₂ ·H ₂ O
Molecular weight:	502.98
Chemical name:	(1S)-1,5-Anhydro-1-C-{4-chloro-3-[{(4-ethoxyphenyl)methyl]phenyl}-D-glucitol mono-(2S)-propane-1,2-diolate monohydrate

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

Review Results

January 6, 2014

[Brand name]	Forxiga 5 mg tablets, Forxiga 10 mg tablets
[Non-proprietary name]	Dapagliflozin Propylene Glycolate Hydrate
[Applicant]	Bristol-Myers K.K.
[Date of application]	March 15, 2013

[Results of review]

Based on the submitted data, it is concluded that the efficacy of the product in patients with type 2 diabetes mellitus has been demonstrated and the safety is acceptable in view of its observed benefits. However, further investigation is still necessary for the impact of concomitant oral hypoglycemic agents on the safety depending on the type and dose; safety on hypoglycaemia, urinary tract infections, genital infections, polyuria/pollakiuria, volume depletion, increase in ketone body, weight decreased, renal disorder, bone metabolism, cardiovascular risks, malignant tumors, etc.; and safety in patients with renal or hepatic impairment and in elderly patients, etc.

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

[Indication]	Type 2 diabetes mellitus
[Dosage and administration]	The usual adult dosage is 5 mg of dapagliflozin administered orally once daily. The dose may be increased to 10 mg once daily for the patient with an inadequate response to the 5 mg dose if the patient is followed up carefully.

Review Report (1)

November 8, 2013

I. Product Submitted for Registration

[Brand name]	Forxiga 5 mg tablets, Forxiga 10 mg tablets
[Non-proprietary name]	Dapagliflozin Propylene Glycolate Hydrate
[Name of applicant]	Bristol-Myers K.K.
[Date of application]	March 15, 2013
[Dosage form/Strength]	Each tablet contains Dapagliflozin Propylene Glycolate Hydrate equivalent to 5 or 10 mg of dapagliflozin.
[Proposed indication]	Type 2 diabetes mellitus
[Proposed dosage and administration]	The usual adult dosage is 5 mg of dapagliflozin administered orally once daily. The dose may be increased to 10 mg once daily for the patient with an inadequate response to the 5 mg dose if the patient is followed up carefully.

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

The data submitted in this application and the outline of 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.

Forxiga 5 mg and Forxiga 10 mg tablets are tablets containing Dapagliflozin Propylene Glycolate Hydrate (hereinafter referred to as "dapagliflozin") as the active ingredient, which is a selective human sodium-glucose co-transporter (SGLT) 2 inhibitor discovered by US Bristol-Myers Squibb Company. SGLT2 is the major glucose co-transporter specifically expressed in the proximal renal tubules and involved in reabsorption of glucose from the glomerular filtrate. Patients with familial renal glycosuria, who have mutations in SGLT2 gene, have been reported to show persistent excretion of glucose in urine (Santer R, et al. *J Am Soc Nephrol*. 2003;14:2873-82, Calado J, et al. *Nephrol Dial Transplant*. 2008;23:3874-9). As described above, selective SGLT2 inhibitors exert hypoglycemic activity in an insulin-independent manner by promoting glucose excretion in urine, thus are unlikely to induce hypoglycaemia.

A regulatory application for the product has recently been submitted by the applicant because its efficacy and safety in patients with type 2 diabetes mellitus have been demonstrated.

As of September 2013, the product has been approved in 36 countries in Europe and other areas, and is currently under review in the US.¹

2. Data relating to quality

2.A *Summary of the submitted data*

2.A.(1) *Drug substance*

2.A.(1.1) *Characterization*

The drug substance is a white to pale yellowish white powder and has been determined for description, melting point, hygroscopicity, crystalline polymorphism, specific rotation, solubility,

¹ An application was submitted in 2010, but most up-to-date safety information (from nonclinical studies and all clinical studies including ongoing clinical studies) was requested in January 2012 for further evaluation of the benefit-risk of the product.

pH, dissociation constant, partition coefficient, X-ray powder diffraction, and particle size distribution.

2.A.(1).2 Manufacturing process

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

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

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

Table 1. Stability studies on drug substance

Study	Primary batches	Temperature	Humidity	Storage form	Storage period
Long term	3 pilot scale batches	5°C	-	Low-density polyethylene bag (double) + high-density polyethylene container	24 months
	3 pilot scale batches	25°C	60% RH		36 months
	3 pilot scale batches	30°C	65% RH		36 months
Accelerated	3 pilot scale batches	40°C	75% RH		6 months

Based on the above, a retest period of [REDACTED] months has been proposed for the drug substance when stored in double polyethylene bags inside a polyethylene container at room temperature.

2.A.(2) Drug product

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

The drug product is immediate release tablets (film-coated tablets) containing 6.150 mg (5 mg of dapagliflozin) or 12.30 mg (10 mg of dapagliflozin) of the drug substance per tablet. It contains microcrystalline cellulose, anhydrous lactose, crospovidone, silicon dioxide, magnesium stearate,

partially hydrolyzed polyvinyl alcohol, titanium oxide, Macrogol 4000, talc, and yellow ferric oxide as excipients.

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

[REDACTED]

[REDACTED].

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

The proposed specifications for the drug product include content, description, identification (IR, liquid chromatography [HPLC]), uniformity of dosage units (content uniformity test [HPLC]), disintegration, water content, and assay.

In the course of the regulatory review, specifications for related substances (HPLC) and dissolution were added.

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

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

Table 2. Stability studies on drug product

Study	Primary batches	Temperature	Humidity	Storage form	Storage period
Long term	3 pilot scale batches	5°C	-	PTP package High-density polyethylene bottle package	36 months
	3 pilot scale batches	25°C	60% RH		36 months
	3 pilot scale batches	30°C	75% RH		36 months
Accelerated	3 pilot scale batches	40°C	75% RH		6 months

Based on the above, a shelf-life of 36 months has been proposed for the drug product when packaged in PTP (polyvinyl chloride/polychloro-trifluoroethylene/aluminum foils) or high-density polyethylene bottles (with desiccant) and stored at room temperature.

2.B Outline of the review by PMDA

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

2.B.(1) Stability of drug product

[REDACTED]

The applicant responded as follows:

Under the stress conditions (25°C/60% RH, unpackaged, 12 months), increases in related substances and water content as well as decreases in disintegration and hardness were observed, but these variabilities fell within the acceptance criteria. Therefore, the drug product was expected

to be stable for a short period of time during storage in the automatic tablet packaging machine or one-dose packages.

PMDA asked the applicant to explain the need to include cautions for storage (e.g., to avoid high temperature and humidity) in handling instructions.

The applicant responded that a caution statement will be included regarding the need to avoid high temperature and humidity after removing the drug product from PTP sheets or bottles.

PMDA accepted the response.

2.B.(2) Dissolution of drug product

PMDA asked the applicant to explain the relationship between dissolution and disintegration of the drug product and the reason for including disintegration testing in the specifications.

The applicant responded as follows:

[REDACTED]

[REDACTED]

[REDACTED]. Based on the above, PMDA instructed the applicant to also include dissolution testing in the specifications for the finished product in order to detect the impact of formulation and/or process changes on the bioavailability of the product etc.

The applicant responded as follows:

[REDACTED].

PMDA accepted the response.

3. Non-clinical data

3.(i) Summary of pharmacology studies

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

As primary pharmacodynamic studies, *in vitro* studies on mechanism of action, and *in vivo* studies on urinary glucose excretion promoting activity and hypoglycemic action were conducted in diabetic and nondiabetic animal models. As secondary pharmacodynamic studies, inhibition of various receptors etc., were investigated. As safety pharmacology studies, effects on the cardiovascular system were investigated. Effects on central nervous and respiratory systems were evaluated in repeated oral dose toxicity studies. Effects on hERG current were evaluated in a non-GLP study.² Pharmacology studies were performed using the free form of Dapagliflozin Propylene Glycolate Hydrate. Also, its dose levels are expressed as free form.

² This study was conducted as a non-GLP study because it was out of scope of “The non-clinical evaluation of the potential for delayed ventricular repolarization (QT-interval prolongation) by human pharmaceuticals” (PFSB/ELD Notification No. 1023-4 dated October 23, 2009) at that time.

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

3.(i).A.(1).1 *In vitro* studies

(a) Inhibition of SGLT2 and SGLT1 by dapagliflozin (4.2.1.1.1 to 4.2.1.1.4)

Inhibition of human sodium-glucose co-transporter (SGLT) 2 and SGLT1 by dapagliflozin was evaluated in human SGLT2- or SGLT1-expressing CHO cells.³ The results showed that the IC₅₀ values (mean ± standard error [SE]) of dapagliflozin against SGLT2 and SGLT1 were 1.12 ± 0.065 and 1391 ± 7 nM, respectively, and the Ki values (mean ± SE) of dapagliflozin for SGLT2 and SGLT1 were 0.55 ± 0.16 and 810 ± 200 nM, respectively. Results of the investigation on mode of inhibition showed a competitive and reversible inhibition of SGLT2 by dapagliflozin. In addition, inhibition of SGLT2 and SGLT1 by dapagliflozin was evaluated in CHO cells expressing rat, mouse, or dog SGLT2 or SGLT1. The results showed that the IC₅₀ values of dapagliflozin against SGLT2 and SGLT1 were 3.0 ± 0.5 and 620 ± 70 nM, respectively, in rats; 2.3 ± 0.6 and 299 ± 166 nM, respectively, in mice; and 1.6 ± 1.0 and 698 ± 203 nM, respectively, in dogs. On the other hand, the IC₅₀ values of phlorizin, a nonselective SGLT inhibitor, against SGLT2 and SGLT1 were 35.6 ± 4.2 and 330 ± 50 nM, respectively, in humans; 75 ± 8 and 302 ± 30 nM, respectively, in rats; 60 ± 22 and 364 ± 239 nM, respectively, in mice; and 51 ± 19 and 357 ± 95 nM, respectively, in dogs.

(b) Inhibition of SGLT2 and SGLT1 by human metabolites of dapagliflozin (4.2.1.1.1, 4.2.1.1.5)

Inhibition of SGLT2 and SGLT1 by desethyl dapagliflozin (a human metabolite of dapagliflozin) was evaluated in CHO cells expressing human and rat SGLT2 or SGLT1.³ The results showed that the IC₅₀ values (mean ± SE) of desethyl dapagliflozin against SGLT2 and SGLT1 were 1.0 ± 0.1 and 1500 ± 100 nM, respectively, in humans and 2.4 ± 0.4 and 260 ± 30 nM, respectively, in rats. On the other hand, the IC₅₀ values of phlorizin against SGLT2 and SGLT1 were 34 ± 6 and 270 ± 22 nM, respectively, in humans, and 75 ± 8 and 302 ± 30 nM, respectively, in rats.

Similarly, inhibition of SGLT2 and SGLT1 by dapagliflozin 3-*O*-glucuronide and dapagliflozin 2-*O*-glucuronide (human metabolites of dapagliflozin) was evaluated.³ The results showed that the IC₅₀ values of dapagliflozin 3-*O*-glucuronide against SGLT2 and SGLT1 were 2900 ± 252 and >80,000 nM, respectively and that the IC₅₀ values of dapagliflozin 2-*O*-glucuronide against SGLT2 and SGLT1 were 4400 ± 356 and >80,000 nM, respectively. On the other hand, the IC₅₀ values of phlorizin against SGLT2 and SGLT1 were 37 ± 69 and 572 ± 169 nM, respectively.

3.(i).A.(1).2 *In vivo* studies

(a) Studies in nondiabetic animals

i) Study in SGLT2-knockout mice (single dose) (4.2.1.1.6)

A single oral dose of dapagliflozin (0.1, 1, 10 mg/kg) or vehicle⁴ was administered to male wild-type mice and SGLT2-knockout (KO) mice (n = 6-9/group) and urine was collected for 3 hours post-dose under food- and water-deprived conditions. Urinary excretions of glucose, sodium (Na), potassium (K), and calcium (Ca) per hour per kg body weight and urine output were measured. As a result, urinary glucose excretion and urine output increased in wild-type mice in a dose-dependent manner and increased significantly in all dose groups of dapagliflozin compared with the control group. Urinary Na excretion increased significantly in the 10 mg/kg group compared with the control group, but no significant difference was observed in urinary K and Ca excretions. In SGLT2-KO mice, urinary glucose excretion increased significantly in the 10 mg/kg group compared with the control group, but no significant difference was observed in urine output and urinary Na, K, and Ca excretions between any dose group of dapagliflozin and the control group.

³ Sodium-dependent uptake of ¹⁴C-labeled α-methylglucopyranoside was used as the measure.

⁴ 1% ethanol solution

ii) Study in normal rats (single dose) (4.2.1.1.7)

A single oral dose of dapagliflozin (0.01, 0.1, 1, 10 mg/kg) or vehicle⁵ was administered to male rats (n = 3/group) under fasted conditions, and then an oral glucose tolerance test (OGTT) was performed with glucose solution (2 g/kg). Feeding was restarted 1 hour after OGTT, and urine was collected under fed conditions for 24 hours post-dose. As a result, urinary glucose excretion for 24 hours post-dose increased significantly in the 1 and 10 mg/kg groups and urine output for 24 hours post-dose increased significantly in the 0.1, 1 and 10 mg/kg groups compared with the baseline values.⁶ Additional male rats (n = 3/group) received administration in the same manner, and blood glucose was measured over time for the first 24 hours after OGTT. The results showed a dose-dependent decrease in blood glucose for 1 hour post-dose and a significant decrease in blood glucose AUC_{0-1h} (mean ± SE) in the 1 and 10 mg/kg groups compared with the control group (75.80 ± 3.55, 60.78 ± 7.93, 48.00 ± 5.25, and 34.98 ± 0.75 mg·h/dL in the dapagliflozin 0.01, 0.1, 1, and 10 mg/kg groups, respectively; 69.11 ± 2.17 mg·h/dL in the control group).

iii) Study in normal rats (duration of urinary glucose excretion promoting activity (single dose) (4.2.1.1.8)

A single oral dose of dapagliflozin 1 mg/kg or vehicle⁵ was administered to male rats (n = 6/group), and urine was collected periodically under fed conditions for 168 hours post-dose. As a result, urinary glucose excretion per hour in the dapagliflozin group was 73 ± 6, 74 ± 5, 12 ± 3, 0.89 ± 0.5, 0.22 ± 0.12, and 0.14 ± 0.12 mg/h during 0 to 6, 0 to 24, 24 to 48, 48 to 72, 72 to 96, and 96 to 168 hours post-dose, respectively; the excretion increased significantly during 0 to 6, 0 to 24, and 24 to 48 hours post-dose compared with the control group (<0.3 mg/h during any time period). Urine output per hour increased significantly in the dapagliflozin group compared with the control group during 0 to 6 and 0 to 24 hours post-dose.

(b) Studies in diabetic animal models

i) Study in streptozotocin-induced diabetic rats (single dose) (4.2.1.1.9)

A single intraperitoneal dose of streptozotocin (STZ) (65 mg/kg) was administered to male rats (n = 5-6/group), and then a single oral dose of dapagliflozin (0.01, 0.03, or 0.1 mg/kg) or vehicle⁵ was administered 4 days after STZ administration. Blood glucose was measured over time under fasted conditions for 5 hours post-dose. As a result, blood glucose decreased in a dose-dependent manner and the decrease was significant between 2 and 5 hours post-dose in the dapagliflozin 0.03 mg/kg group and between 1 and 5 hours post-dose in the 0.1 mg/kg group compared with the control group.

ii) Studies in ZDF rats (single dose) (4.2.1.1.10, 4.2.1.1.12)

A single oral dose of dapagliflozin (0.01, 0.1, 1, 10 mg/kg) or vehicle⁵ was administered to male ZDF rats (19 weeks of age, n = 6/group), and measurements of blood glucose over time and urine collection were performed for 24 hours post-dose. The study was performed under fasted conditions between 0 and 6 hours post-dose, and under fed conditions between 6 and 24 hours post-dose. The results showed significant increases in urinary glucose excretion and urine output during 0 to 6 hours post-dose in all dose groups of dapagliflozin compared with the control group. Blood glucose at 6 hours post-dose decreased significantly in all dose groups of dapagliflozin compared with the control group. Three of 6 animals in the 10 mg/kg group died between 6 and 24 hours post-dose; neither food nor water consumption was observed during 6 to 24 hours post-dose in these animals⁷ [see “3.(i).B.(3) Effect of volume depletion”].

⁵ 20 mmol/L sodium diphosphate solution containing 5% N-methyl-2-pyrrolidone and 20% polyethylene glycol 400

⁶ Baseline values were obtained from the urine collected in metabolic cages for 18 hours pre-dose under fasted conditions.

⁷ The applicant explained that, based on the results of the follow-up studies (4.2.1.1.11) to explore the causes of death observed in this study, the deaths in the 3 animals were caused by dehydration resulting from inability to compensate with adequate food and water intake for the fasting effects and the increased urine output resulting from the pharmacological activity because of stress coming from housing in a metabolic cage.

In addition, another study was performed, where a single oral dose of dapagliflozin (0.01, 0.1, or 1 mg/kg) or vehicle⁵ was administered to male ZDF rats (17 weeks of age, n = 6/group) and blood glucose was measured over time for 24 hours post-dose. The study was performed under fasted conditions between 0 and 6 hours post-dose, and under fed conditions between 6 and 24 hours post-dose. The results showed that blood glucose decreased significantly in the 0.01 mg/kg group between 4 and 24 hours post-dose and in the 0.1 and 1 mg/kg groups between 2 and 24 hours post-dose compared with the control group (blood glucose [mean ± SE] at 24 hours post-dose was 378 ± 16, 385 ± 17, and 272 ± 12 mg/dL in the dapagliflozin 0.01, 0.1, and 1 mg/kg groups, respectively, and 454 ± 11 mg/dL in the control group).

iii) Study in ZDF rats (repeated dose) (4.2.1.1.13)

Dapagliflozin (0.01, 0.1, 1, 10 mg/kg) or vehicle⁵ was orally administered once daily for 15 days to male ZDF rats (17 weeks of age, n = 6/group). Urine was collected for 24 hours on Days 2 and 6 under fed conditions and on Day 14 under fasted conditions, and blood glucose was measured on Days 8 and 15 under fasted conditions and on Day 14 under fed conditions just prior to dosing. As a result, urinary glucose excretion and urine output were not affected on Days 2 to 3 and Days 6 to 7 (under fed conditions) in any dose group of dapagliflozin compared with the control group, but increased dose-dependently on Days 14 to 15 (under fasted conditions) and showed a significant increase in the ≥0.1 mg/kg groups compared with the control group. Blood glucose decreased dose-dependently on Days 8 and 15 (under fasted conditions) and on Day 14 (under fed conditions), and decreased significantly in all dose groups of dapagliflozin compared with the control group except for on Day 15 in the 0.01 mg/kg group. One of 6 animals in the 10 mg/kg group died 24 hours after the last dose.⁸

iv) Hyperinsulinemic-euglycemic clamp study in ZDF rats (repeated dose) (4.2.1.1.14)

Dapagliflozin 0.5 mg/kg or vehicle⁵ was orally administered once daily for 15 days to male ZDF rats (15 weeks of age, n = 6/group), and hyperinsulinemic-euglycemic clamp⁹ for 120 minutes was performed at 48 hours after the last dose. As a result, the glucose infusion rate (GIR) (mean ± SE) was 6.0 ± 0.6 in the dapagliflozin group and 2.6 ± 0.4 mg/kg/min in the control group, showing a significant increase in the dapagliflozin group compared with the control group. The glucose utilization rate¹⁰ (mean ± SE) was 6.6 ± 0.32 in the dapagliflozin group and 5.3 ± 0.15 mg/kg/min in the control group, showing a significant increase in the dapagliflozin group compared with the control group. The endogenous glucose production rate¹¹ (mean ± SE) was 0.7 ± 0.4 in the dapagliflozin group and 3.0 ± 0.32 mg/kg/min in the control group, showing a significant decrease in the dapagliflozin group compared with the control group. In addition, a bolus of ¹⁴C-labeled D-glucose was administered 90 minutes after the start of insulin administration, and the tissue glucose uptake was evaluated in the skeletal muscle, fatty tissue, and liver isolated after the completion of the study. The results showed that the glucose uptake in the liver increased significantly in the dapagliflozin group compared with the control group, while no effects were observed in the skeletal muscle and fatty tissue.

⁸ The applicant explained that the reason for the death is unknown.

⁹ Following administration of human insulin (genetical recombination) at 38.7 mU/kg/min for 10 minutes, continuous infusion was started at 20 mU/kg/min, and 10% unlabeled glucose solution was infused so that the blood glucose level is maintained at 120 mg/dL. ³H-labeled D-glucose was continuously administered from 60 minutes before the start of insulin administration until the end of treatment.

¹⁰ (Endogenous glucose utilization) - (urinary glucose excretion rate). The glucose disposal rate (mg/kg/min) was calculated by dividing the rate of ³H-labeled D-glucose infusion (dpm/kg/min) by the ³H-labeled D-glucose specific activity (dpm/mL) at plasma glucose concentrations (mg/mL).

¹¹ (Glucose disposal rate) - GIR

(c) Studies in obese animal models

i) Study in obese ZDF rats (hypoglycemic activity) (repeated dose) (4.2.1.1.17)

Male ZDF rats (6 weeks of age, n = 8/group) were treated orally with dapagliflozin 1 mg/kg, rosiglitazone¹² 10 mg/kg, or vehicle⁵ once daily for 5 weeks and ZDF lean rats (6 weeks of age, n = 6) were treated orally with vehicle⁵ once daily for 5 weeks, and an OGTT was performed 24 hours after the last dose (after an overnight fast). The results showed that AUC of change in blood glucose (mean ± SE) over 180 minutes after OGTT in ZDF rats was 11,938 ± 1014, 17,526 ± 808, and 32,847 ± 3995 mg·min/dL in the dapagliflozin, rosiglitazone, and control groups, respectively, showing a significant decrease in the dapagliflozin and rosiglitazone groups compared with the control group. The change (mean ± SE) in plasma insulin concentration at 30 minutes after OGTT was 7.2 ± 1.2, 3.3 ± 0.9, and 1.1 ± 0.4 ng/mL in the dapagliflozin, rosiglitazone, and control groups, respectively, showing a significant increase in the dapagliflozin group compared with the control and rosiglitazone groups. In ZDF lean rats, AUC of change in blood glucose was 8317 ± 504 mg·min/dL, and the change in plasma insulin concentration was 0.9 ± 0.3 ng/mL.

Additional male ZDF rats and ZDF lean rats (6 weeks of age, n = 5/group) received administration in the same manner, and HbA1c was determined 5 weeks after administration. The results showed a significant decrease in HbA1c in the dapagliflozin and rosiglitazone groups compared with the control group.

ii) Hyperinsulinemic-euglycemic clamp study in obese ZDF rats (repeated dose) (4.2.1.1.18)

Male ZDF rats (6-7 weeks of age, n = 7-10/group) were treated orally with dapagliflozin 1 mg/kg, rosiglitazone 10 mg/kg, or vehicle⁵ once daily for 5 weeks and ZDF lean rats (6-7 weeks of age, n = 7) were treated orally with vehicle⁵ once daily for 5 weeks, and hyperinsulinemic-euglycemic clamp for 90 minutes was performed 24 hours after the last dose (after an overnight fast).¹³ The results showed that the GIR (mean ± SE) in ZDF rats increased significantly in the dapagliflozin group compared with the control group (the GIR in ZDF rats was 28 ± 1, 30 ± 1, and 21 ± 2 mg/kg/min in the dapagliflozin, rosiglitazone, and control groups, respectively, and that in control ZDF lean rats was 50 ± 3 mg/kg/min).

iii) Study in obese ZDF rats (effects on glucose and fatty acid metabolisms) (repeated dose) (4.2.1.1.19)

Male ZDF rats (7 weeks of age, n = 13-17/group) were treated orally with dapagliflozin 0.5 mg/kg, rosiglitazone¹² 10 mg/kg, or vehicle¹⁴ once daily for 5 weeks and ZDF lean rats (7 weeks of age, n = 12) were treated orally with vehicle¹⁴ once daily for 5 weeks, and HbA1c, blood glucose, and plasma insulin concentrations were measured over time. From some animals (n = 6/group¹⁵), urine was collected from 48 to 72 hours post-dose after 5 weeks of treatment, and urine output and urinary glucose excretion were measured. Additional animals (n = 5/group) received administration in the same manner, and haematology was performed under fed or overnight fasted conditions at pre-dose and 48 hours post-dose after 1 and 5 weeks of treatment. Furthermore, additional animals (n = 5/group) received administration in the same manner, and hepatic glycogen, hepatic triglycerides (TGs), and pancreatic insulin content were measured in the liver and pancreas isolated under fed or overnight fasted conditions 48 hours after the last dose. The results¹⁶ showed that urine output and urinary glucose excretion in ZDF rats 48 to 72 hours after the last dose decreased significantly in the dapagliflozin group compared with the control group.

¹² The free form of rosiglitazone was used, and its dose levels are expressed as free form equivalents.

¹³ Following administration of human insulin (genetical recombination) at 38.7 mU/kg/min for 10 minutes, continuous infusion was started at 20 mU/kg/min, and 20% glucose solution was infused so that the blood glucose level is maintained at 130 mg/dL.

¹⁴ Distilled water was used as the vehicle for dapagliflozin, and 0.5% carboxymethylcellulose was used as the vehicle for rosiglitazone. Distilled water was administered to control ZDF and ZDF lean rats.

¹⁵ A total of 0 of 6 animals in the dapagliflozin group, 4 of 6 animals in the rosiglitazone group, 4 of 6 control ZDF rats, and 3 of 6 control ZDF lean rats were the same animals used in the evaluation of HbA1c etc.

¹⁶ Results from the rosiglitazone group are not shown.

In the control group, HbA1c and blood glucose increased throughout the treatment period, but in the dapagliflozin group, these values were significantly decreased from 1 week after the start of treatment through the treatment period compared with the control group, demonstrating an inhibition of blood glucose elevation associated with disease progression (HbA1c [mean \pm SE] at baseline and after 5 weeks of treatment in ZDF rats were $3.9\% \pm 0.0\%$ and $5.4\% \pm 0.1\%$ in the dapagliflozin group and $3.9\% \pm 0.1\%$ and $9.7\% \pm 0.4\%$ in the control group, and those in control ZDF lean rats were $3.6\% \pm 0.0\%$ and $4.3\% \pm 0.1\%$, respectively). In the dapagliflozin group, plasma insulin concentrations remained at the baseline values during 2 weeks of treatment but increased significantly thereafter compared with the control group.¹⁷ Body weight increased significantly in the dapagliflozin group compared with the control group, but no effects were observed on food consumption. An evaluation of nonesterified fatty acids (NEFA), TG, aspartate aminotransferase (AST), alanine aminotransferase (ALT), and haematocrit values after 5 weeks of treatment revealed a significant decrease in ALT in the dapagliflozin group compared with the control group (under fed conditions). The pancreatic insulin level under fasted conditions after 5 weeks of treatment increased significantly in the dapagliflozin group compared with the control group, but no effects were observed under fed conditions. No differences were observed in hepatic glycogen and hepatic TGs between the dapagliflozin and control groups both under fasted and fed conditions.

iv) Hyperglycaemic clamp study in obese ZDF rats (repeated dose) (4.2.1.1.15, 4.2.1.1.16)

Female ZDF rats (7 weeks of age, n = 14/group) were treated orally with dapagliflozin 1 mg/kg or vehicle¹⁸ from the first day of the high fat diet loading and ZDF lean rats were treated with vehicle from the first day of the normal diet feeding once daily for 34 days.¹⁹ Hyperglycaemic clamp for 90 minutes was performed 48 hours after the last dose (after an overnight fast).²⁰ The results showed that the insulin sensitivity index (M/I index; i.e., GIR divided by plasma insulin concentration) and the pancreatic β -cell disposition index (DI; i.e., plasma C-peptide concentration multiplied by M/I index) decreased significantly in control ZDF rats compared with control ZDF lean rats, and those in ZDF rats increased significantly in the dapagliflozin group compared with the control group. Pancreatic sections were prepared at 48 hours after the last dose (after an overnight fast) from about half of the animals, and no effects of dapagliflozin were observed on the percentage of insulin-stained pancreatic β -cell area. However, the percentage of the area of pancreatic β -cells densely stained with insulin and the pancreatic islet morphology (as measured by pancreatic β -cell area divided by the β -cell cluster number) were significantly improved in the dapagliflozin group compared with the control group.

In addition, female ZDF rats (7 weeks of age, n = 14/group) were treated orally with dapagliflozin (1 mg/kg) or vehicle¹⁸ from 10 days after the start of the high fat diet loading and ZDF lean rats were treated with vehicle from 10 days after the start of the normal diet feeding once daily for 34 days.²¹ Hyperglycaemic clamp for 90 minutes was similarly performed 48 hours after the last dose (after an overnight fast). The results showed that the M/I index and DI decreased significantly in control ZDF rats compared with control ZDF lean rats, and those in ZDF rats increased significantly in the dapagliflozin group compared with the control group. Pancreatic sections were prepared 48 hours after the last dose (after an overnight fast) from about half of the

¹⁷ Plasma insulin concentrations in the control ZDF rats increased compared with those in the control ZDF lean rats, peaked at Week 1 of the treatment, and decreased over time. Plasma insulin levels in obese ZDF rats are higher than those in ZDF lean rats due to the increased insulin secretion for compensation for peripheral insulin resistance, but characteristically decline depending on the progression of hyperglycaemia (Finegood, et al. *Diabetes*. 2001;50:1021-9).

¹⁸ Distilled water

¹⁹ Blood glucose (mean \pm SE) at the start of treatment was 108 ± 5 , 108 ± 4 , and 98 ± 2 mg/dL in the dapagliflozin, control, and ZDF lean groups, respectively.

²⁰ Following administration of glucose 375 mg/kg, 25% glucose solution was infused so that the blood glucose level is maintained at >97.2 mg/dL.

²¹ Blood glucose (mean \pm SE) at the start of treatment was 133 ± 7 , 135 ± 7 , and 101 ± 2 mg/dL in the dapagliflozin, control, and ZDF lean groups, respectively.

ZDF rats, and no effects of dapagliflozin were observed on the pancreatic β -cell area. However, the percentage of the area of pancreatic β -cells densely stained with insulin and the pancreatic islet morphology were significantly improved in the dapagliflozin group compared with the control group.

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

3.(i).A.(2).1 *In vitro* studies

(a) Inhibition for other SGLT isoforms (4.2.1.1.1, 4.2.1.2.1)

Inhibitions of human SGLT4, SGLT6, and sodium-myoinositol co-transporter (SMIT) 1 by dapagliflozin were evaluated in CHO cells expressing these SGLT isoforms.²² As a result, the K_i values (mean \pm SE) for SGLT4, SGLT6, and SMIT1 were 3.3 ± 0.7 , 0.80 ± 0.1 , and $14 \pm 2 \mu\text{M}$, respectively. In a similar evaluation of rat SMIT1, the IC_{50} value was $5.2 \pm 0.26 \mu\text{M}$.

(b) Inhibition for glucose transporters (4.2.1.1.1, 4.2.1.1.5, 4.2.1.2.2)

Inhibitions by dapagliflozin (20, 50, 100 μM), phloretin (20 μM), and cytochalasin B (20 μM) were evaluated in human erythrocytes expressing glucose transporter (GLUT) 1, human liver cancer-derived HepG2 cells expressing GLUT2, and human differentiated adipocytes expressing GLUT4.²³ As a result, 100 μM dapagliflozin inhibited GLUT1 by $3.6\% \pm 3.6\%$ (mean \pm SE), GLUT2 by $11.6\% \pm 3.2\%$, and GLUT4 by $33\% \pm 4\%$. Phloretin and cytochalasin B inhibited GLUT1 by $4.6\% \pm 3.9\%$ and $47.6\% \pm 12.4\%$, respectively, GLUT2 by $53.6\% \pm 5.4\%$ and $86.2\% \pm 4.7\%$, respectively, and GLUT4 by $44\% \pm 7\%$ (phloretin only).

Inhibitions²⁴ by dapagliflozin (20 μM) or cytochalasin B (20 μM) under insulin stimulated or non-insulin stimulated conditions were evaluated in primary human adipocytes. The results showed that the inhibition rates (mean \pm SE) under non-insulin stimulated and insulin stimulated conditions by dapagliflozin were $9\% \pm 1\%$ and $8\% \pm 3\%$, respectively, and those by cytochalasin B were $88\% \pm 2\%$ and $89\% \pm 0.3\%$, respectively. In a similar evaluation in mouse 3T3-L1 adipocytes, the inhibition rates by dapagliflozin were $20\% \pm 4\%$ and $19\% \pm 7\%$, respectively, and those by cytochalasin B were $93\% \pm 3\%$ and $92\% \pm 0.3\%$, respectively.

In addition, inhibitions²⁴ by dapagliflozin 3-*O*-glucuronide (a human metabolite of dapagliflozin) (20, 100, 250, 500 μM), phloretin (20 μM), and cytochalasin B (20 μM) under insulin stimulated or non-insulin stimulated conditions were evaluated in primary human adipocytes. The results showed that the inhibition rates under non-insulin stimulated and insulin stimulated conditions by dapagliflozin 3-*O*-glucuronide (500 μM) were $26\% \pm 12\%$ and $42\% \pm 6\%$, respectively, those by phloretin were $71\% \pm 7\%$ and $69\% \pm 6\%$, respectively, and those by cytochalasin B were $92\% \pm 2\%$ and $93\% \pm 2\%$, respectively.

(c) Inhibition for various receptors, ion channels, transporters, and enzymes (4.2.1.2.5, 4.2.1.2.3 [Reference data]; 4.2.1.2.6, 4.2.1.2.7)

Inhibitions of 286 different receptors, ion channels, transporters, or enzymes by 10 μM dapagliflozin were evaluated. As a result, no inhibitions of $\geq 50\%$ were detected. Inhibitions of human calcitonin receptor and vitamin D receptor by 10 μM dapagliflozin were also evaluated. As a result, no inhibition was detected for any of these receptors. In addition, inhibitions of 329 different receptors, ion channels, transporters, or enzymes by 10 μM dapagliflozin 3-*O*-glucuronide (a major human metabolite of dapagliflozin) were evaluated. As a result, no inhibitions of $\geq 50\%$ were detected. Furthermore, inhibitions of 40 different receptors, ion

²² Inhibition of SGLT4 was measured by sodium-dependent uptake of ^{14}C -labeled α -methylglucopyranoside, and inhibitions of SGLT6 and SMIT1 were measured by sodium-dependent uptake of ^3H -labeled myoinositol.

²³ Inhibition of GLUT1 was measured by uptake of ^3H -labeled glucose, inhibition of GLUT2 was measured by uptake of ^3H -labeled deoxy-D-glucose, and inhibition of GLUT4 was measured by uptake of ^3H -labeled deoxy-D-glucose under insulin stimulation.

²⁴ Uptake of ^{14}C -labeled deoxy-D-glucose was used as the measure.

channels, or enzymes by 30 µM desethyl dapagliflozin (an active metabolite of dapagliflozin) were evaluated. As a result, no inhibitions of ≥50% were detected.

3.(i).A.(2).2 *In vivo studies*

(a) Effect on endogenous glucose production (single dose) (4.2.1.2.8)

A single oral dose of dapagliflozin (0.5, 1.0 mg/kg) or vehicle¹⁸ was administered to male ZDF rats (11 weeks of age, n = 6-8/group) and ZDF lean rats (11 weeks of age, n = 6-8/group), and blood glucose and plasma insulin concentrations were measured over time and urine was collected from 0 to 60 minutes post-dose and from 60 to 120 minutes post-dose. In addition, ³H-labeled glucose was continuously administered from 60 minutes pre-dose to 120 minutes post-dose of dapagliflozin, and the endogenous glucose production rate²⁵ was determined. The results showed that, in ZDF rats, urinary glucose excretion increased significantly from 0 to 60 minutes post-dose and from 60 to 120 minutes post-dose and blood glucose also decreased significantly from 30 to 120 minutes post-dose in all dose groups of dapagliflozin compared with the control group. However, in ZDF lean rats, though urinary glucose excretion increased significantly from 60 to 120 minutes post-dose in all dose groups of dapagliflozin compared with the control group, no significant changes were found in blood glucose except for a significant decrease in the 0.5 mg/kg group at 2 hours post-dose. The endogenous glucose production rate increased significantly in the 1 mg/kg group compared with the control group both in ZDF and ZDF lean rats.

(b) Effects on body weight and body composition (repeated dose) (4.2.1.2.10)

Dapagliflozin (0.5, 1, 5 mg/kg), a cannabinoid type 1 receptor antagonist (positive control, 10 mg/kg), or vehicle⁵ was administered orally once daily for 27 days under ad libitum feeding to male rats²⁶ (n = 8/group) fed with the high fat diet or high carbohydrate diet for past 10 weeks. Similarly, dapagliflozin (5 mg/kg) or vehicle⁵ was administered orally once daily for 27 days under restricted feeding.²⁷ Body composition was measured by MRI at baseline and on Day 22, and clinical chemistry was performed on Day 27 (after an overnight fast). The results showed that, under ad libitum feeding, water consumption, urine output, and urinary glucose excretion increased significantly in all dose groups of dapagliflozin compared with the control group. Food consumption tended to increase in all dose groups of dapagliflozin compared with the control group, but body weight on Day 25 decreased from baseline by 3.9%, 4.2%, and 5.6% in animals treated with dapagliflozin 0.5, 1, and 5 mg/kg, respectively.²⁸ In positive control animals, a body weight reduction by 24.0% from baseline was observed associated with the marked decrease in food consumption. Regarding body composition, fat mass decreased significantly (as change from baseline) in animals treated with dapagliflozin 0.5 and 5 mg/kg compared with control animals, while no significant differences were observed in lean mass (as change from baseline).²⁹ Fatty acid metabolism was enhanced, as evidenced by significant increases in 3-β-hydroxybutyric acid and NEFA in all dose groups of dapagliflozin and the significant increase in glycerol in the 5 mg/kg group.³⁰ Also, a significant increase in blood urea nitrogen (BUN) was observed compared with the control group. The fasting blood glucose levels on Day 27 decreased significantly in all dose groups of dapagliflozin compared with the control group.

²⁵ Calculated by dividing the rate of ³H-labeled D-glucose infusion (dpm/kg/min) by the ³H-labeled D-glucose specific activity (dpm/mL) at plasma glucose concentrations (mg/mL).

²⁶ Age was unknown.

²⁷ Food consumption was restricted so that daily food consumption is identical between the control (restricted diet) group and the 5 mg/kg (ad libitum feeding) group, and between the 5 mg/kg (restricted diet) group and the vehicle (ad libitum feeding) group.

²⁸ Body weight decreased from baseline by 12.3% in the 5 mg/kg group and 3.9% in the control group under restricted feeding.

²⁹ Fat mass (as change from baseline) and body water decreased significantly in the 5 mg/kg group under restricted feeding compared with the control group.

³⁰ In the 5 mg/kg group under restricted feeding, 3-β-hydroxybutyric acid, NEFA, and glycerol increased significantly and fasting blood glucose decreased significantly compared with the control group.

3.(i).A.(2).3 Studies on potential relationship with bladder cancer

(a) Impact on gene expression (repeated dose) (4.2.1.2.11)

Dapagliflozin 0.5 mg/kg or vehicle¹⁸ was administered orally once daily for 5 weeks to male ZDF rats (7 weeks of age, n = 5/group), and alterations in gene expression were examined using microarrays in cells isolated from the liver, skeletal muscle, kidneys, and fatty tissue 48 hours after the last dose (under fasted or fed conditions). The results showed no alterations in the expression of cell proliferation-related genes.

(b) Effect of glucose on the proliferation of bladder cancer cell lines (4.2.1.2.12)

Effect of glucose (at concentrations of 11, 25, 35, and 50 mM) on the cell proliferation rate was investigated using 5 bladder cancer cell lines (T-24, TCCSUP, UM-UC-3, J82, SW780). The results showed that the cell proliferation rate was not increased by addition of high concentration glucose in any cell line.

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

3.(i).A.(3).1 Effects on central nervous system (4.2.3.2.5, 4.2.3.2.8, 4.2.3.7.7.2)

Animals in the dapagliflozin 150 mg/kg/day and control³¹ groups in the 6-month repeated oral dose toxicity study in rats were subjected to an examination for general symptoms and a neuroelectrophysiological evaluation³² after 6 months of treatment (4.2.3.2.5). In addition, animals in the dapagliflozin 120 mg/kg/day and control³¹ groups in the 12-month repeated oral dose toxicity study in dogs were subjected to a neuroelectrophysiological evaluation³² after 6 and 12 months of treatment (4.2.3.2.8). As a result, no neurotoxic effects related to dapagliflozin were confirmed in either rats or dogs, and behavioral changes evaluated as part of general symptoms were not observed in rats or dogs until the dose increased to 25 mg/kg/day or 120 mg/kg/day, respectively. Regarding plasma exposure to dapagliflozin, C_{max} and AUC_{0-24h} in rats treated at 25 mg/kg/day were 42.1 µg/mL and 314 µg·h/mL, respectively, which are equivalent to approximately 220-fold the plasma C_{max} and 432-fold the plasma AUC_τ,³³ respectively, at the maximum recommended clinical dose. C_{max} and AUC_{0-24h} in dogs treated at 120 mg/kg/day were 167 µg/mL and 1540 µg·h/mL, respectively, which are equivalent to approximately 874-fold the plasma C_{max} and 2118-fold the plasma AUC_τ, respectively, at the maximum recommended clinical dose. Animals in the dapagliflozin 150 mg/kg/day and control³¹ groups in the 3-month repeated oral dose toxicity study in rats (4.2.3.7.7.2) were subjected to an electroencephalographic evaluation after 11 weeks of treatment. As a result, no effects of dapagliflozin were observed, and C_{max} and AUC_{0-24h} in animals treated at 150 mg/kg/day were 79.4 µg/mL and 797.5 µg·h/mL, respectively, which are equivalent to approximately 416-fold C_{max} and 1097-fold AUC_τ,³³ respectively, at the maximum recommended clinical dose.

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

(a) In vitro study (4.2.1.3.1)

Effects of dapagliflozin (10, 30 µM)³⁴ on hERG currents were evaluated in HEK293 cells expressing hERG channels. The results showed that the inhibition rates of hERG currents relative to baseline (mean ± SE) by dapagliflozin at concentrations of 10 and 30 µM were 3.7% ± 2.0% and 15% ± 5.1%, respectively. In addition, effects of dapagliflozin (3, 10, 30 µM) on action potential parameters (resting membrane potential, overshoot, maximal upstroke velocity, action potential durations at 50% and 90% repolarization) were evaluated in rabbit Purkinje fibers. The results showed no effects of up to 30 µM dapagliflozin on any parameter compared with findings

³¹ 90% polyethylene glycol solution

³² Performed as a non-GLP evaluation.

³³ The plasma C_{max} (191 ng/mL) and AUC_τ (727 ng·h/mL) of unchanged dapagliflozin on Day 14 observed in a clinical pharmacology study (Study MB102025; 5.3.3.2.1), in which dapagliflozin was orally administered once daily for 14 days in Japanese patients with type 2 diabetes mellitus at the maximum recommended clinical dose (10 mg/day). By assuming the percent plasma protein binding value in humans to be 91.0% (4.2.2.3.1), the unbound concentrations were calculated.

³⁴ 0.03% dimethylsulfoxide (vehicle)

in the vehicle³⁴ group. The concentration of dapagliflozin 30 µM is equivalent to approximately 714-fold the unbound plasma C_{max}³³ of dapagliflozin at the maximum recommended clinical dose.

(b) In vivo study (4.2.1.3.2)

To conscious dogs (n = 3/sex), a single oral dose of vehicle³¹ was administered, and 2 days later, a single oral dose of dapagliflozin (30 mg/kg) was administered. The heart rate, left ventricular pressure, systemic arterial pressure (systolic and diastolic blood pressures, mean blood pressure), electrocardiographic parameters (including RR, PR, QRS, and QT/QT₈₀ intervals³⁵), and spontaneous locomotor activity were evaluated at 1 hour pre-dose and at approximately 20 hours post-dose. The results showed no effects of dapagliflozin on any parameter. The plasma C_{max} and AUC_{0-24h} of dapagliflozin at 30 mg/kg were estimated to be 51.7 µg/mL and 597 µg·h/mL, respectively,³⁶ which are equivalent to approximately 271-fold the plasma C_{max} and 821-fold the plasma AUC_τ,³³ respectively, at the maximum recommended clinical dose.

3.(i).A.(3).3 Effects on respiratory system (4.2.3.2.4 to 4.2.3.2.8)

Animals in the 3- and 6-month repeated oral dose toxicity studies in rats were evaluated for effects on respiratory status as part of general symptoms. The results of the 3- and 6-month studies showed no effects of dapagliflozin up to 50 mg/kg/day and 25 mg/kg/day, respectively (4.2.3.2.4, 4.2.3.2.5). Also, animals in the 3- and 12-month repeated oral dose toxicity studies in dogs were evaluated for effects on respiratory status. The results of the 3- and 12-month studies showed no effects of dapagliflozin up to 180 mg/kg/day and 120 mg/kg/day, respectively (4.2.3.2.7, 4.2.3.2.8). In addition, no effects of dapagliflozin on the arterial oxygen saturation were observed up to 250 mg/kg/day in the 1-month repeated oral dose toxicity study in dogs (4.2.3.2.6).

Regarding plasma exposure of dapagliflozin, C_{max} and AUC_{0-24h} in rats treated at 50 mg/kg/day were 58.4 µg/mL and 438 µg·h/mL, respectively, which are equivalent to approximately 306-fold the plasma C_{max} and 602-fold the plasma AUC_τ,³³ respectively, at the maximum recommended clinical dose. C_{max} and AUC_{0-24h} in dogs treated at 250 mg/kg/day were 196.7 µg/mL and 2667.5 µg·h/mL, respectively, which are equivalent to approximately 1030-fold the plasma C_{max} and 3669-fold the plasma AUC_τ,³³ respectively, at the maximum recommended clinical dose.

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

3.(i).B.(1) Mechanism of action

PMDA asked the applicant to explain the pharmacological activity of dapagliflozin in humans in light of the biological distribution, functions, and homology with SGLT2 of each SGLT isoform as well as selectivity of dapagliflozin for SGLT2, etc.

The applicant responded as follows:

Human SGLT2 has been reported to be a glucose transporter selectively expressed in the kidneys,³⁷ and SGLT2 is highly expressed in the kidneys also in mice, rats, and dogs.³⁸ Although SGLT2 binding site(s) in dapagliflozin has not been identified, amino acid sequences of rat, mouse, and dog SGLT2 exhibit 91% to 96% homology with that of human SGLT2,³⁹ and the IC₅₀ values (mean ± SE) of dapagliflozin against SGLT2 in humans, rats, mice, and dogs are 1.12 ± 0.065, 3.0 ± 0.5, 2.3 ± 0.6, and 1.6 ± 1.0 nM, respectively, indicating comparable inhibition

³⁵ Defined as QT interval individually corrected with a correction factor obtained by using a standardized heart rate (80 bpm) (Miyazaki H, et al. *Exp Anim.* 2002;51:465-475).

³⁶ C_{max} (males, 42.0 µg/mL; females, 61.3 µg/mL) and AUC_{0-24h} (males, 560 µg·h/mL; females, 634 µg·h/mL) on Day 1 in the 3-month repeated oral dose toxicity study in dogs (4.2.3.2.7).

³⁷ Chen, et al. *Diabetes Ther.* 2010;1:57-92, Tazawa, et al. *Life Sci.* 2005;76:1039-50

³⁸ You, et al. *J Biol Chem.* 1995;270:29365-71, Toyono T, et al. *Cell Tissue Res.* 2011;345:243-52, Affymetrix Inc. HT_MG-430_PM Array (for mice), Vallon V, et al. *J Am Soc Nephrol.* 2011;22:104-12, Affymetrix Inc. RAE230_A Array (for rats), Affymetrix Inc. Canine Genome 2.0 Array Canine_2 (for dogs).

³⁹ Data obtained by using AlignX program of bioinformatics software Vector NTI based on the total amino acid sequence in the Homologene database at NCBI (as of June 14, 2013).

potencies among these species (4.2.1.1.1, 4.2.1.1.3, 4.2.1.1.4). In addition, based on the findings in patients with familial renal glycosuria who have mutations in SGLT2 gene and in SGLT2-KO mice etc., SGLT2 has been reported to have a function of glucose reabsorption in the renal tubules.⁴⁰ Based on the above, SGLT2 is considered to show no species-specific differences in terms of the distribution and functions.

Human SGLT1 has been reported to be highly expressed in the small intestine, skeletal muscle, and heart,³⁷ and it has been suggested that SGLT1 is selectively and highly expressed in the gastrointestinal tract, kidneys, and thyroid gland in mice, in the small intestine in rats, and in the jejunum in dogs.³⁸ SGLT1 has been believed to be primarily involved in the active transport of glucose and galactose via the apical membrane.⁴⁰ Human SGLT1 has been reported to be expressed also in the kidneys, and it shows 58% homology with SGLT2.³⁹ The unbound plasma concentration of dapagliflozin (42 nM) at the maximum recommended clinical dose in humans corresponded to 76-fold the Ki value of dapagliflozin for human SGLT2 and 0.05-fold the Ki value for SGLT1, suggesting that dapagliflozin would primarily inhibit SGLT2 in humans. On the other hand, in SGLT2-KO mice, the significant increase in urinary glucose excretion observed in the dapagliflozin 10 mg/kg group suggested that dapagliflozin at 10 mg/kg would lead to an SGLT2-independent increase in urinary glucose excretion (4.2.1.1.6). The unbound plasma concentration of dapagliflozin at a dose of 10 mg/kg in mice was estimated to be 945 nM,⁴¹ which corresponds to approximately 3-fold the IC₅₀ value (299 nM) of dapagliflozin against mouse SGLT1, suggesting that dapagliflozin would exhibit a urinary glucose excretion promoting activity through inhibition of SGLT1. The unbound plasma concentrations of dapagliflozin at doses of 0.1, 1, and 10 mg/kg in rats were estimated to be 7.2, 72, and 720 nM, respectively (4.2.1.1.7), which correspond to 2.4-, 24-, and 240-fold the IC₅₀ value (3.0 nM) of dapagliflozin against rat SGLT2 and 0.01-, 0.12-, and 1.2-fold the IC₅₀ value (620 nM), respectively, against rat SGLT1, suggesting that pharmacological activity of dapagliflozin at 10 mg/kg in rats may be partly mediated by inhibition of SGLT1. Dapagliflozin was 1242 times more selective to SGLT2 than to SGLT1 in humans; this factor was higher than that in rats (207 times), mice (130 times), and dogs (436 times) (4.2.1.1.1, 4.2.1.1.3, 4.2.1.1.4).

Reported functions of other SGLT isoforms include the following: glucose sensor, which is primarily expressed in the small intestine and skeletal muscle (SGLT3); transporter of mannose, glucose, or fructose, which is primarily expressed in the small intestine and skeletal muscle (SGLT4); transporter of mannose, glucose, or fructose, which is primarily expressed in the kidneys (SGLT5); and transporter of myo-inositol or glucose, which is ubiquitously expressed (SGLT6, SMIT1).⁴⁰ The primary physiological roles of SGLT3 to 6 and SMIT have not been adequately reported, but they show 45% to 56% homology with human SGLT2, with no major difference in the homology among dogs, mice, and rats.³⁹ Dapagliflozin was 210 to 190,000 times more selective to SGLT2 than to SGLT3 to 6 and SMIT1 in humans, showing a high selectivity for SGLT2.⁴²

Based on the above, dapagliflozin is considered to exert hypoglycemic activity in humans through urinary glucose excretion promoting activity by selectively inhibiting SGLT2.

PMDA accepted the response because dapagliflozin has been confirmed to be more selective for SGLT2 than for the other isoforms studied, although SGLT isoforms with unknown specific characteristics (including function) still exist.

⁴⁰ Wright EM, et al. *Mol Aspects Med.* 2013;34:183-96, Santer R, et al. *Clin J Am Soc Nephrol.* 2010;5:133-41, Grempler R, et al. *FEBS Lett.* 2012;586:248-53

⁴¹ Estimated by assuming linearity using the unbound plasma C_{max} (2200 nM) of dapagliflozin observed on Day 1 of treatment with dapagliflozin 4.1 mg/kg in the 1-week repeated oral dose toxicity study in mice (4.2.3.2.1) (plasma protein binding in mice, 92.8%; [4.2.2.3.1]).

⁴² Suzuki M, et al. *J Pharmacol Exp Ther.* 2012;341:692-701

3.(i).B.(2) Duration of action

PMDA asked the applicant to explain the duration of action of dapagliflozin.

The applicant responded as follows:

In normal rats treated with a single dose (under fed conditions) of dapagliflozin 1 mg/kg (4.2.1.1.8), urinary glucose excretion per hour increased significantly compared with the control rats during 0 to 6, 0 to 24, and 24 to 48 hours post-dose. Urinary glucose excretion per hour during 0 to 6 hours post-dose was similar to that during 0 to 24 hours post-dose (73 and 74 mg/h, respectively), and that during 24 to 48 hours post-dose was as low as 12 mg/h; thus, dapagliflozin is expected to exert urinary glucose excretion promoting activity primarily during 0 to 24 hours post-dose. In ZDF rats treated with a single dose of dapagliflozin 1 mg/kg (4.2.1.1.10), urinary glucose excretion per hour increased significantly compared with the control rats during 0 to 6 (under fasted conditions) and 0 to 24 (under fed conditions from 6 hours post-dose) hours post-dose (348.8 and 415.3 mg/h, respectively). Although a rigorous comparison cannot be made due to reasons such as differences in the pathology and feeding conditions, the urinary glucose excretion promoting activity for the first 24 hours post-dose was observed also in ZDF rats.

Following a single oral dose of dapagliflozin 1 and 10 mg/kg in ZDF rats, the unbound plasma concentration of dapagliflozin peaked at approximately 5 hours post-dose, and C_{max} was estimated to be 61 and 627 nM, respectively,⁴³ which were approximately 20- and 200-fold the IC₅₀ (3.0 nM) of dapagliflozin, respectively, against rat SGLT2. Following administration of dapagliflozin 1 and 10 mg/kg, urinary glucose excretion during 0 to 6 hours post-dose (1.85 and 1.82 g, respectively) increased significantly compared with the control (0.11 g), and in addition, blood glucose at 6 hours post-dose (111 and 86 mg/dL, respectively) decreased significantly compared with the control (313 mg/dL). Also, blood glucose at 24 hours post-dose of dapagliflozin 1 and 10 mg/kg (225.0 and 151.6 mg/dL, respectively) significantly decreased compared with the control (341.2 mg/dL). The unbound plasma concentrations of dapagliflozin at 24 hours post-dose were estimated to be 4 and 42.3 nM, respectively,⁴³ which were similar to or above the IC₅₀ value (3.0 nM) of dapagliflozin against rat SGLT2. Persistence of hypoglycemic activity up to 24 hours post-dose was demonstrated based on the finding that blood glucose at 24 hours post-dose was significantly low after dosing dapagliflozin at ≥ 0.1 mg/kg compared with the control. Furthermore, in obese ZDF rats treated orally with dapagliflozin 1 mg/kg once daily for 5 weeks (4.2.1.1.17), HbA1c after 5 weeks of treatment (4.6%) was significantly low compared with that in the control rats (7.8%); thus, effectiveness of once daily administration of dapagliflozin was demonstrated.

PMDA accepted the applicant's response [see "4.(iii).B.(5).1 Dosage regimen" for justification for the clinical dosage regimen].

3.(i).B.(3) Effect of volume depletion

The applicant explained as follows:

Dapagliflozin promotes urinary glucose excretion, which causes osmotic diuresis resulting in increased urine output and volume depletion. In a single-dose study in ZDF rats (4.2.1.1.10), of 6 animals in the dapagliflozin 10 mg/kg group, 3 animals that had consumed no food or water between 6 and 24 hours post-dose died. Thus, 2 follow-up studies were conducted in order to explore the factors (4.2.1.1.11). There were multiple stressors in the previous study including the use of metabolic cages, fasting for 6 hours post-dose, and periodical blood collection up to 24 hours post-dose. The first follow-up study explored these factors as potential causes of the deaths.

⁴³ C_{max} of dapagliflozin at doses of 1 and 10 mg/kg was 1240 and 12,800 nM, respectively, and the plasma concentrations of dapagliflozin at 24 hours post-dose were below the lower limit of quantitation and 863 nM, respectively. The plasma concentration of dapagliflozin at 24 hours after administration of dapagliflozin 1 mg/kg was estimated by assuming linearity. By assuming the plasma protein binding in rats as 95.1% (4.2.2.3.1), unbound concentrations were calculated.

Male ZDF rats (17-19 weeks of age, n = 6/group) kept in metabolic cages were treated with a single oral dose of dapagliflozin 10 mg/kg or vehicle⁵ under the following conditions: (a) fasting for 6 hours post-dose (fasted group), (b) repeated blood collection (blood collection group), or (c) fasting for 6 hours post-dose plus repeated blood collection (fasted + blood collection group). The results showed that 3 animals in the fasted group, 5 animals in the blood collection group, and 3 animals in the fasted + blood collection group became moribund or died within 48 hours after administration of dapagliflozin. However, no animals became moribund or died in the control group. In the moribund or dead animals, decreases in food and water consumption and a trend toward increased body fluid loss⁴⁴ were observed at 24 hours post-dose. On the other hand, no death during the treatment period occurred in animals treated orally with dapagliflozin 10 mg/kg once daily for 14 days in a normal cage, demonstrating tolerability under this condition (4.2.1.1.13). In the second follow-up study, male ZDF rats (17-19 weeks of age, n = 6/group) kept in metabolic cages were treated with a single oral dose of dapagliflozin 10 mg/kg or vehicle,⁵ and were subjected to serum chemistry and pathological examinations at 6 and 24 hours post-dose (under fasted conditions for 6 hours post-dose). The results showed that decreased food consumption during 6 to 24 hours post-dose, decreased water consumption during 0 to 24 hours post-dose, and an increased body fluid loss were observed in some animals treated with dapagliflozin, and these animals exhibited a trend toward increased serum levels of β -hydroxybutyric acid and urea nitrogen and decreased serum levels of bicarbonate. Therefore, the moribund or dead animals were considered to have increased serum urea nitrogen levels (due to increased body fluid loss and decreased renal perfusion) and worsening metabolic ketoacidosis (due to decreased food consumption and increased urinary glucose excretion).

Based on the above, the moribundity or death seen in ZDF rats treated with dapagliflozin 10 mg/kg may have been caused by inability to compensate with adequate food and water intake for the fasting effects and the increased urine output and urinary glucose excretion resulting from the pharmacological activity because of the stress coming from housing in a metabolic cage. The plasma C_{max} and AUC_{0-24h} of dapagliflozin at 10 mg/kg were estimated to be 5.235 $\mu\text{g}/\text{mL}$ and 49.318 $\mu\text{g}\cdot\text{h}/\text{mL}$, respectively, which are equivalent to approximately 27-fold the plasma C_{max} and 68-fold the plasma AUC,³³ respectively, at the maximum recommended clinical dose.

PMDA accepted the applicant's explanation [see "4.(iii).B.(3).5) Volume depletion" for safety in humans].

3.(ii) Summary of pharmacokinetic studies

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

The pharmacokinetics of single intra-arterial, intravenous, and oral doses of dapagliflozin or ¹⁴C-dapagliflozin in rats, dogs, and monkeys were evaluated. In addition, the repeat-dose pharmacokinetics was evaluated based on toxicokinetics observed in repeat-dose toxicity studies in mice, rats, and dogs. Furthermore, the distribution was evaluated in rats; metabolism and excretion were evaluated in mice, rats, and dogs; and placental transfer and excretion in milk were evaluated in rats. The plasma concentrations of dapagliflozin and its metabolites (desethyl dapagliflozin, dapagliflozin 3-O-glucuronide) were determined by high performance liquid chromatography/tandem mass spectrometry (LC-MS/MS). The lower limit of quantitation of dapagliflozin was 39 nM (16 ng/mL) and that of desethyl dapagliflozin was 39 nM (16 ng/mL) in rat, dog, and monkey plasma, and the lower limit of quantitation was 50 ng/mL in mouse plasma. Measurement of radioactivity in biological samples was performed using whole-body autoradiography and liquid scintillation counting. Primary study results are shown below. Pharmacokinetic studies were performed using dapagliflozin. Its dose levels are expressed as free form (dapagliflozin).

⁴⁴ Defined as the difference between urine output and water consumption.

3.(ii).A.(1) Absorption (4.2.2.2.1, 4.2.3.2.1 to 4.2.3.2.8)

The pharmacokinetic parameters of unchanged dapagliflozin following a single intra-arterial, intravenous, or oral dose under fasted conditions⁴⁵ in male rats, dogs, and monkeys (n = 3/species) were as shown in Table 3.

Table 3. Pharmacokinetic parameters of unchanged dapagliflozin after a single dose of dapagliflozin

Species (number of animals)	Route of administration	Dose (mg/kg)	C _{max} (μ g/mL)	AUC _{inf} (μ g·h/mL)	t _{max} (h)	t _{1/2} (h)	CL _p (mL/min/kg)	V _{ss} (L/kg)	BA (%)
Rat (3)	i.a.	1	-	3.55 ± 0.42	-	4.6 ± 0.8	4.8 ± 0.6	1.6 ± 0.1	-
	p.o.	1	0.60 ± 0.46	2.96 ± 0.73	1.7 ± 2.0	NC	-	-	84 ± 21
Dog (3)	i.v.	6.6	-	76.4 ± 10.1	-	7.4 ± 1.2	1.5 ± 0.2	0.8 ± 0.1	-
	p.o.	6.6	10.7 ± 1.6	63.6 ± 7.3	0.6 ± 0.4	NC	-	-	83 ± 2
Monkey (3)	i.v.	6	-	17.1 ± 6.8	-	3.5 ± 1.9	6.4 ± 2.3	0.8 ± 0.2	-
	p.o.	6	1.54 ± 0.40	4.27 ± 2.17	1.9 ± 1.8	NC	-	-	25 ± 2

Mean ± standard deviation (SD); -, Not applicable; NC, Not calculated

i.a., Intra-arterial; p.o., Per oral; i.v., Intravenous

C_{max}, Maximum plasma concentration; AUC_{inf}, Area under the plasma concentration-time curve from time 0 to infinity; t_{max}, Time to reach the maximum plasma drug concentration; t_{1/2}, Apparent terminal phase elimination half-life; CL_p, Total plasma clearance; V_{ss}, Volume of distribution; BA, Absolute bioavailability.

With repeated oral administration of dapagliflozin at a dose of 4.1, 25, 43, or 75 mg/kg/day once daily for 1 week in male and female mice (n = 3/group/timepoint), at a dose of 5, 50, or 300 mg/kg/day once daily for 1 month in male and female rats (n = 3/group/timepoint), and at a dose of 5, 25, or 250 mg/kg/day once daily for 1 month in male and female dogs (n = 3/group), the concentrations of unchanged dapagliflozin are roughly liner. The accumulation ratio calculated from C_{max} in female rats was 1.9, while those calculated from AUC_{0-t} and C_{max} in male rats and male and female dogs were in the range of 1.17 to 1.36.

3.(ii).A.(2) Distribution (4.2.2.2.1, 4.2.2.3.1 to 4.2.2.3.4)

Following a single oral dose of ¹⁴C-dapagliflozin 22 mg/kg⁴⁶ in male and female rats (n = 1/timepoint) under fasted conditions, the tissue radioactivity levels⁴⁷ peaked within 4 hours post-dose. The tissue to blood ratio of radioactivity concentrations based on AUC_{inf} was high in the cecum (8.72 in males, 9.29 in females), large intestine (8.58, 15.2), kidneys (4.96, 4.59), renal cortex (6.83, 6.21), renal medulla (3.15, 3.23), small intestine (4.59, 3.79), liver (4.34, 3.92), bile in the bile duct (8.45, 5.15), Harderian gland (4.84, 9.70), adrenal gland (3.42, 3.61), salivary gland (3.28, 5.48), and brown fat (2.02, 3.30) and <3 in the remaining tissues. The ratio was low in the bone (0.134, 0.106) and eye (crystalline lens) (0.158, 0.075), and the brain to blood ratio was 0.250 in males and 0.335 in females. The tissue radioactivity levels declined gradually throughout the study period, and fell below the lower limit of quantitation in the majority of tissues at 72 hours post-dose, with radioactivity detectable only in the Harderian gland and kidneys at 168 hours post-dose. Following a single oral dose of ¹⁴C-dapagliflozin 26.6 mg/kg in male rats (n = 3/timepoint) under fasted conditions, elimination of radioactivity⁴⁸ from pigmented skin was more rapid than that from non-pigmented skin (t_{1/2} was 80.7 hours in non-pigmented skin, 3.4 hours in pigmented skin). In other tissues, the elimination tended to be generally slower compared with that seen in the tissue distribution study using whole-body autoradiography, but no major differences were observed.

Following a single oral dose of ¹⁴C-dapagliflozin 23 mg/kg in pregnant rats (Gestation day 18, n = 1/timepoint) under fasted conditions, dapagliflozin crossed the placenta, and tissue

⁴⁵ Dapagliflozin was administered intra-arterially or intravenously over 10 minutes. The crossover design was used for dogs and monkeys.

⁴⁶ The target dose was 25 mg/kg, but the actual average dose was approximately 22 mg/kg in each group.

⁴⁷ Based on whole-body autoradiography

⁴⁸ Based on liquid scintillation counting

radioactivity⁴⁷ was detected in the fetuses from 30 minutes post-dose. The tissue radioactivity levels in the maternal animals and fetuses peaked within 4 to 8 hours post-dose in the majority of tissues, and the tissue to blood ratios of radioactivity concentrations based on AUC_{inf} in the uterus, vagina,⁴⁹ placenta, fetal intestine, fetal liver, fetal kidneys, fetal brain, and fetal blood were 3.09, 2.09, 1.25, 1.29, 1.13, 0.876, 0.795, and 0.642, respectively. The tissue radioactivity levels in the fetal liver, kidneys, brain, and blood fell below the lower limit of quantitation at ≥72 hours post-dose, while the radioactivity in the fetal intestine remained even at 96 hours post-dose.

Following a single oral dose of ¹⁴C-dapagliflozin 5.2 mg/kg in lactating rats (Postpartum day 8 or 9, n = 3/timepoint), radioactivity⁴⁸ was detected in the milk from 30 minutes post-dose and peaked at 2 hours post-dose. The milk to plasma ratios of radioactivity concentrations based on C_{max} and AUC_{inf} were 0.554 and 0.762, respectively.

Pooled fresh blood of rats, dogs, and monkeys containing dapagliflozin (10 µM) was incubated and its distribution in blood cells was investigated. As a result, the mean distribution in red blood cells was 10% to 23%. The mean plasma protein binding (equilibrium dialysis method) of dapagliflozin at 0.5 and 5 µg/mL in mice, rats, rabbits, and dogs was in the range of 93% to 95%, and that of dapagliflozin 3-O-glucuronide at 0.5 and 5 µg/mL was in the range of 91% to 95%.

3.(ii).A.(3) Metabolism (4.2.2.2.1, 4.2.2.4.1 to 4.2.2.4.3)

Following a single oral dose of ¹⁴C-dapagliflozine 200 mg/kg in male mice (n = 5/timepoint), unchanged dapagliflozine in urine and feces accounted for 22.4% (10.3% in urine, 12.1% in feces) and metabolites in urine and feces accounted for 53.6% (27.2% in urine, 26.4% in feces) of the administered radioactivity at 120 hours post-dose; oxidative metabolites accounted for approximately 47% and glucuronidated metabolites accounted for approximately 6% of the administered dose. Urinary metabolites that accounted for ≥3% of the administered radioactivity included a mixture of desethyl dapagliflozin, hydroxy-dapagliflozin-2, and dapagliflozin carboxylic acid (14.5%) and benzylic hydroxy-dapagliflozin (3.38%). Fecal metabolites that accounted for ≥3% of the administered radioactivity included a mixture of desethyl dapagliflozin, hydroxy-dapagliflozin-2, and dapagliflozin carboxylic acid (9.75%), hydroxy-dapagliflozin-3 (6.51%), and benzylic hydroxy-dapagliflozin (5.85%). In plasma, unchanged dapagliflozin accounted for 65% and metabolites accounted for 23.8% of the total plasma radioactivity (AUC_{0-t}), and metabolites that accounted for ≥3% of the total plasma radioactivity included dapagliflozin O-glucuronide (8.0%), benzylic hydroxy-dapagliflozin (5.2%), and a mixture of desethyl dapagliflozin, hydroxy-dapagliflozin-2, and dapagliflozin carboxylic acid (4.4%).

Following a single oral dose of ¹⁴C-dapagliflozin 25 mg/kg in male rats (n = 3/timepoint), unchanged dapagliflozin in urine and feces accounted for 25.4% (14.6% in urine, 10.8% in feces) and metabolites in urine and feces accounted for 51.7% (21.4% in urine, 30.3% in feces) of the administered radioactivity at 168 hours post-dose; oxidative metabolites accounted for approximately 51% and glucuronidated metabolites accounted for approximately 1% of the administered dose. Urinary metabolites that accounted for ≥3% of the administered radioactivity included a mixture of desethyl dapagliflozin and hydroxy-dapagliflozin-2 (9.1%), desethyl dapagliflozin glucuronide-1 (5.3%), and a mixture of benzylic hydroxy-dapagliflozin and dapagliflozin O-glucuronide (3.6%). Fecal metabolites that accounted for ≥3% of the administered radioactivity included desethyl dapagliflozin (19.3%), oxo-dapagliflozin-3 (4.2%), and benzylic hydroxy-dapagliflozin (3.6%). In plasma, unchanged dapagliflozin accounted for 84.9% and metabolites accounted for 10.4% of the plasma radioactivity (AUC_{0-24h}), and the

⁴⁹ The animals were planned to be sacrificed at 168 hours post-dose, but were euthanized after delivery at 96 hours post-dose, because delivery occurred earlier than expected. For the vagina, pharmacokinetic parameters were calculated excluding the data from rats euthanized at 96 hours post-dose.

metabolite that accounted for $\geq 3\%$ of the plasma radioactivity was benzylic hydroxy-dapagliflozin (3.7%).

Following a single oral dose of ^{14}C -dapagliflozin 20 mg/kg in male bile-duct-cannulated rats ($n = 3$), unchanged dapagliflozin in urine and bile accounted for 14.5% and 2.0%, respectively, of the administered radioactivity and metabolites in urine and bile accounted for 31.9% and 25.0%, respectively, of the administered radioactivity at 24 hours post-dose. Urinary metabolites that accounted for $\geq 3\%$ of the administered radioactivity included desethyl dapagliflozin glucuronide-1 (9.1%), desethyl dapagliflozin (5.4%), and dapagliflozin 3-*O*-glucuronide (4.6%). Biliary metabolites that accounted for $\geq 3\%$ of the administered radioactivity included dapagliflozin 3-*O*-glucuronide (6.7%) and desethyl dapagliflozin glucuronide-1 (5.4%).

Following a single oral dose of ^{14}C -dapagliflozin 25 mg/kg in male dogs ($n = 3$), unchanged dapagliflozin in urine and feces accounted for 43.4% (6.4% in urine, 37.0% in feces) and metabolites in urine and feces accounted for 42.4% (13.9% in urine, 28.5% in feces) of the administered radioactivity at 168 hours post-dose; oxidative metabolites accounted for approximately 41% and glucuronidated metabolites accounted for approximately 2% of the administered dose. Urinary metabolites that accounted for $\geq 2\%$ of the administered radioactivity included desethyl dapagliflozin glucuronide-1 (3.3%), oxo-dapagliflozin-3 (2.6%), and desethyl dapagliflozin glucuronide-3 (2.13%). Fecal metabolites that accounted for $\geq 2\%$ of the administered radioactivity included oxo-dapagliflozin-3 (11.1%), dapagliflozin carboxylic acid (7.6%), and desethyl dapagliflozin (6.9%). In plasma, unchanged dapagliflozin accounted for 84.7% and metabolites accounted for 15.5% of the plasma radioactivity ($\text{AUC}_{0-12\text{h}}$), and metabolites that accounted for $\geq 2\%$ of the plasma radioactivity included oxo-dapagliflozin-3 (4.0%), dapagliflozin 3-*O*-glucuronide (2.5%), dapagliflozin 3-*O*-glucuronide (2.1%), and desethyl dapagliflozin glucuronide-1 (2.0%).

In the *in vivo* studies in mice, rats, and dogs using ^{14}C -dapagliflozin, a number of trace amounts of metabolites (≥ 16 metabolites in mice, ≥ 14 metabolites in rats, ≥ 11 metabolites in dogs) were detected in addition to the major metabolite.

Rat, dog, and monkey liver microsomes were incubated with dapagliflozin in the presence of 3 μM NADPH or 10 μM UDPGA. As a result, the metabolic rates of dapagliflozin via oxidative metabolism and glucuronidation were found to be highest in rats (100 and 40 pmol/min/mg protein via oxidative metabolism and glucuronidation, respectively), followed in descending order by monkeys (90 and 30 pmol/min/mg protein, respectively) and dogs (70 and 10 pmol/min/mg protein, respectively). After an incubation of rat, dog, and monkey hepatocytes with dapagliflozin (3 μM), no metabolic activities were found in rats, while the metabolic rates in dogs and monkeys were 39 and 24 pmol/min/million cells, respectively.

As a result of determination of metabolites of ^{14}C -dapagliflozin using mouse, rat, dog, and monkey liver microsomes and hepatocytes, 7 different⁵⁰ metabolites including dapagliflozin 3-*O*-glucuronide were identified. The predominant metabolite(s) were benzylic hydroxy-dapagliflozin in mouse and monkey liver microsomes; hydroxy-dapagliflozin-3 in rat liver microsomes and rat, dog, and monkey hepatocytes; and hydroxy-dapagliflozin-3 and the desethyl dapagliflozin in dog liver microsomes and mouse hepatocytes. No glutathione adducts were detected in hepatocytes in any species.

⁵⁰ Hydroxy-dapagliflozin-1, hydroxy-dapagliflozin-3, benzylic hydroxy-dapagliflozin, desethyl dapagliflozin, desethyl dapagliflozin glucuronide-1, dapagliflozin 3-*O*-glucuronide, and dapagliflozin 2-*O*-glucuronide. Identification was not attempted for a number of other small radioactivity peaks.

3.(ii).A.(4) Excretion (4.2.2.4.3, 4.2.2.5.1 to 4.2.2.5.3)

Following a single oral dose of ^{14}C -dapagliflozin 200 mg/kg in male mice ($n = 5$), 39.2% and 41.0% of the administered radioactivity were recovered in urine and feces within 120 hours post-dose, respectively. Of the total combined urinary and fecal excretion, 73% and 93% were excreted within 24 and 48 hours post-dose, respectively.

Following a single oral dose of ^{14}C -dapagliflozin 26 mg/kg in male rats ($n = 3$), 39.8% and 49.0% of the administered radioactivity were recovered in urine and feces within 168 hours post-dose, respectively. Of the total combined urinary and fecal excretion, 80% and 97% were excreted within 24 and 48 hours post-dose, respectively.

Following a single oral dose of ^{14}C -dapagliflozin 20 mg/kg in male bile-duct-cannulated rats ($n = 3$), 46.4%, 3.8%, and 27.0% of the administered radioactivity were excreted in urine, feces, and bile within 24 hours post-dose, respectively. Unchanged dapagliflozin in bile accounted for 2% of the administered radioactivity. Following a single oral dose of ^{14}C -dapagliflozin 25 mg/kg in male rats ($n = 3/\text{timepoint}$), unchanged dapagliflozin in feces accounted for 10.8% of the administered radioactivity at 168 hours post-dose, while the value was decreased to 3.8% by bile duct ligation, suggesting that unchanged dapagliflozin excreted in feces was mostly derived from biliary excretion.

Following a single oral dose of ^{14}C -dapagliflozin 24 mg/kg in male dogs ($n = 3$), 21.6% and 72.3% of the administered radioactivity were recovered in urine and feces within 168 hours post-dose, respectively. Of the total combined urinary and fecal excretion, 61% and 91% were excreted within 24 and 48 hours post-dose, respectively.

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

Biological distribution of SGLT2 and its impact on dapagliflozin-related effects

PMDA asked the applicant to explain the biological distribution of SGLT isoforms including SGLT2 and the dapagliflozin-related effects in each tissue.

In humans, SGLT2 is distributed in the renal cortex, SGLT5 in the kidneys, and SMIT in the renal medulla at high levels. In the tissue distribution study in rats using the combustion method and whole-body autoradiography, high radioactivity level was found to be retained for a long time in the kidneys (especially renal medulla).

SGLT1, SGLT3, or SGLT4 is distributed in the human small intestine at high levels; in the tissue distribution study, high radioactivity levels were detected in the gastrointestinal tract including the small and large intestines. In the tissue distribution study using autoradiography, elimination of radioactivity from the small intestine was determined to be relatively rapid, but elimination from the large intestine was relatively slow. Diarrhoea and loose stool observed in animals in the high dose groups (rats, ≥ 1924 -fold AUC at the maximum recommended clinical dose; dogs, ≥ 2091 -fold AUC at the maximum recommended clinical dose) in repeated oral dose toxicity studies were considered related to reduced glucose absorption associated with intestinal SGLT1 inhibition. Therefore, given the difference in the extent of inhibition⁵¹ by dapagliflozin between SGLT2 and SGLT1, SGLT1 inhibition would be more likely to occur in rats than in humans.

SGLT1 is distributed in the human heart at high levels; in tissue distribution studies, higher radioactivity levels were detected in the heart than those in blood. The elimination of radioactivity was determined to be rapid in the tissue distribution study using autoradiography, while the elimination was relatively slow in the tissue distribution study using the combustion method.

⁵¹ Inhibitory activities expressed as IC_{50} against SGLT2 and SGLT1 are approximately 3 and 620 nM in rats and approximately 1 and 1391 nM in humans, respectively.

SMIT is disproportionately distributed in the human thyroid gland at high levels; in tissue distribution studies, relatively high radioactivity levels were detected in the thyroid gland, but the elimination of radioactivity from the thyroid gland was determined to be rapid as in the case of that from blood.

SMIT is disproportionately distributed in the human testes at high levels; in tissue distribution studies, the radioactivity levels in the testes were determined to be relatively low, and the elimination of radioactivity was determined to be slow by autoradiography, while rapid by the combustion method.

No distribution of SGLT2 or other SGLT isoforms has been observed in the human liver. However, in tissue distribution studies, relatively high radioactivity levels were detected in the liver, and the elimination of radioactivity was determined to be rapid by autoradiography, while slow by the combustion method.

No distribution of SGLT2 or other SGLT isoforms has been observed in the human bladder. However, in tissue distribution studies, relatively high radioactivity levels were detected in the bladder of male rats, and the elimination of radioactivity was determined to be rapid. Bacterial flocculation in the bladder and hydronephrosis were observed in the 1-month repeated oral dose toxicity study in rats (4.2.3.2.3) and were considered to be an indirect result of the pharmacological activity, because these changes were consistent with ascending urinary tract infection. In addition, signs that were consistent with urinary tract infection, including sporadic inflammation of the bladder and urothelial hyperplasia, were observed in the 12-month repeated oral dose toxicity study in dogs (4.2.3.2.8), but were not considered to be a direct result of administration of dapagliflozin, because neither dose-relationship nor increase over time was observed in the incidence or severity of these signs. Bladder cancer was not observed in carcinogenicity studies of dapagliflozin in mice and rats.

Among the above tissues, toxicological findings or laboratory abnormalities were also observed in the kidneys, heart, testes, and liver in repeated oral dose toxicity studies, but the exposures at studied dose levels were higher than that at the maximum recommended clinical dose and no impact was observed on the reproductive performance, these findings were not considered to suggest any clinical safety concerns. No toxicological findings were found in the thyroid gland.

Among the above tissues, adverse events involving the gastrointestinal tract (small intestine), thyroid gland, and bladder were reported by subjects who received dapagliflozin in the Japanese phase II study (Study D1692C00005) and a Japanese phase III study (Study D1692C00006), but a causal relationship to dapagliflozin was ruled out for all events except those involving the bladder,⁵² and no safety concerns were raised in the other tissues.

Based on the above, there are no issues requiring special attention regarding the safety of dapagliflozin in tissues where SGLT2 or other SGLTs are distributed at high levels or where high radioactivity levels were detected.

PMDA accepted the applicant's response because the elimination trend in each individual tissue was similar across the studies and time-dependent elimination was seen in all tissues in spite of the observed variability in elimination time according to the approach used in tissue distribution studies. However, toxicological findings and safety in humans will be additionally reviewed in

⁵² Adverse events for which a causal relationship to the study drug could not be ruled out included cystitis and cystitis bacterial, but both were mild in severity.

the toxicology and clinical sections [see “3.(iii) Summary of toxicology studies” and “4.(iii).B.(3) Safety”].

3.(iii) Summary of toxicology studies

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

Toxicity studies of dapagliflozin conducted include single-dose toxicity, repeat-dose toxicity, genotoxicity, carcinogenicity, reproductive and developmental toxicity, and other toxicity studies (mechanistic studies of toxicity, studies on metabolites, etc.). Unless otherwise specified, 90% polyethylene glycol solution was used as the vehicle. Toxicity studies were performed using dapagliflozin unless otherwise specified. Its dose levels are expressed as free form (dapagliflozin).

3.(iii).A.(1) *Single-dose toxicity*

3.(iii).A.(1).1 *Single oral dose toxicity study in mice (4.2.3.1.1)*

A single oral dose of vehicle or dapagliflozin 375, 750, 1500, or 3000 mg/kg was administered to male and female CD-1 mice. Animals in the 3000 mg/kg group died at 3 to 48 hours post-dose (6 of 10 animals [4 of 5 males, 2 of 5 females]). Decrease in locomotor activity and hunched position were observed in animals treated at \geq 1500 mg/kg; and decreased body weight was observed in animals treated at 3000 mg/kg. Based on the above, the approximate lethal dose in mice was determined to be 3000 mg/kg.

3.(iii).A.(1).2 *Single oral dose toxicity study in rats (4.2.3.1.2)*

A single oral dose of vehicle or dapagliflozin 375, 750, 1500, or 3000 mg/kg was administered to male and female SD rats. Animals in the \geq 750 mg/kg groups died on Days 2 to 12 (2 of 10 animals [1 of 5 males, 1 of 5 females] in the 750 mg/kg group; 6 of 10 animals [2 of 5 males, 4 of 5 females] in the 1500 mg/kg group; and 8 of 10 animals [3 of 5 males, 5 of 5 females] in the 3000 mg/kg group). Decrease in locomotor activity, stained fur, reddish nasal discharge, loose stool, and decreased body weight was observed in animals treated at \geq 750 mg/kg; and hunched position was observed in animals treated at \geq 1500 mg/kg. Based on these findings, the approximate lethal dose in rats was determined to be 750 mg/kg.

3.(iii).A.(1).3 *Single oral dose toxicity study in dogs (4.2.3.1.3)*

A single oral dose of vehicle or dapagliflozin 200, 500, or 1000 mg/kg (in 2 divided doses 4 hours apart) were administered to female beagle dogs (n = 3/group). Vomiting was observed at 10 to 60 minutes post-dose. Based on the above, the approximate lethal dose in dogs was determined to be >1000 mg/kg.

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

Repeated oral dose studies of dapagliflozin in mice (1-week, 3-month), rats (1-month, 3-month, 6-month), and dogs (1-month, 3-month, 12-month) were conducted. The primary target organs of toxicity in rats were the kidneys (e.g., dilatation, mineralization, necrosis, and hyperplasia of the renal tubules, hyperplasia of the collecting tubule epithelium and urothelium, exacerbation of chronic nephropathy), bone (increased trabecular bone volume, increased area of ossification), and blood vessel (mineralization in tissues including the kidneys, heart, mammary gland, and intestinal mucosa). In dogs, an increased incidence of vomiting was observed. The plasma exposures (AUC_{0-24h}) at the no observed adverse effect level (NOAEL) in mice (150 mg/kg in the 3-month study), rats (25 mg/kg in the 6-month study), and dogs (120 mg/kg in the 12-month study) were \geq 400-fold, \geq 220-fold, and \geq 2000-fold, respectively, the plasma exposure³³ (AUC_{0-24h}) at the maximum recommended clinical dose.

3.(iii).A.(2).1 One-week repeated oral dose toxicity study in mice (4.2.3.2.1)

Vehicle or dapagliflozin at doses of 4.1, 25, 43, and 75 mg/kg was administered orally to male and female CD-1 mice once daily for 1 week.⁵³ The plasma exposure (AUC_{0-24h}) in females was approximately 2.4 to 4 times higher than that in males, and the exposure (AUC^{54}) after 1 week of treatment in animals treated at 75 mg/kg (96 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males, 298 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females) was ≥ 130 -fold the plasma exposure⁵³ (AUC_{0-24h}) at the maximum recommended clinical dose.

3.(iii).A.(2).2 Three-month repeated oral dose toxicity study in mice (4.2.3.2.2)

Vehicle or dapagliflozin at doses of 50, 150, 250, and 400 mg/kg was administered orally to male and female CD-1 mice once daily for 3 months.⁵⁵ Death⁵⁶ occurred in animals treated at ≥ 250 mg/kg (9 of 20 animals [4 of 10 males, 5 of 10 females] in the 250 mg/kg group; 11 of 20 animals [7 of 10 males, 4 of 10 females] in the 400 mg/kg group). A trend toward increased food consumption, a trend toward increased body weight gain, decreased locomotor activity, abdominal distension, hunched position, abnormal fur, and unkempt fur were observed in animals treated at ≥ 50 mg/kg; and a decrease in the absolute weight of the prostate was observed in animals treated at ≥ 150 mg/kg. Since no changes were observed on necropsy or histopathological examination in animals treated at ≤ 150 mg/kg, the NOAEL was determined to be 150 mg/kg/day.

3.(iii).A.(2).3 One-month repeated oral dose toxicity study in rats (4.2.3.2.3)

Vehicle or dapagliflozin at doses of 5, 50, and 300 mg/kg was administered orally to male and female SD rats once daily for 1 month. Death or moribundity occurred in animals treated at 300 mg/kg (2 of 20 animals [2 of 10 females]). Increases in food and water consumption, urinary glucose, and urine output, decreased urine osmolarity, and increased kidney weight or a trend toward increased kidney weight were observed in animals treated at ≥ 5 mg/kg; increased serum ALT was observed in animals treated at ≥ 50 mg/kg; increased urinary Ca, worsening of clinical conditions accompanied by changes in hematology or clinical chemistry⁵⁷ (including abdominal distension, diarrhoea, rales, dyspnoea, and unkempt fur), decreases in thymus, prostate, and vesicular gland weights, decreased thymic lymphocyte counts, degeneration and mineralization of the glandular stomach, and effects on the kidneys (dilatation of the renal tubules, multifocal medullary tubular necrosis accompanied by mineralization, regenerative hyperplasia, oedema, exacerbation of chronic nephropathy) were observed in animals treated at 300 mg/kg. The applicant explained that the increased serum ALT observed in animals treated at ≥ 50 mg/kg was an adaptive response to the enhanced gluconeogenesis. The applicant also explained that the other findings observed in animals treated at ≤ 50 mg/kg were changes related to the pharmacological activity of dapagliflozin. Based on the above, the NOAEL was determined to be 50 mg/kg/day.

3.(iii).A.(2).4 Three-month repeated oral dose toxicity study in rats (4.2.3.2.4)

Vehicle or dapagliflozin at doses of 5, 50, and 200 mg/kg was administered orally to male and female SD rats once daily for 3 months (with a reversibility assessment after a 1-month recovery period). Death occurred in an animal treated at 200 mg/kg (1 of 30 animals [1 of 15 females]). Increases in food and water consumptions, urinary glucose, urinary Na, urine output, urinary protein, and urinary creatinine, decreased urine osmolarity, increases in serum AST, ALT, and alkaline phosphatase (ALP), etc.,⁵⁸ increased kidney weight, and hypertrophy of collecting tubular epithelial cells were observed in animals treated at ≥ 5 mg/kg; increased urinary Ca, decreased serum protein, decreases in prostate and vesicular gland weights, increased adrenal

⁵³ Evaluation in this study was limited to occurrence of death and toxicokinetics.

⁵⁴ In males, calculated as exposure up to 8 hours post-dose because the plasma concentrations of dapagliflozin at 24 hours post-dose on Day 7 were lower than the detection limit (AUC_{0-8h} for males, AUC_{0-24h} for females).

⁵⁵ Urinalysis was not included in the laboratory tests in the main study.

⁵⁶ A total of 3 of 10 males and 1 of 10 females in the 400 mg/kg group were treated at approximately 10 times the correct dose (4000 mg/kg) on Day 25 due to misdosing, and death of the 3 male animals was found on the next day. Survived animals in this group (6 of 10 females) were euthanized on Day 28.

⁵⁷ Including increases in neutrophil and monocyte counts, and increases in serum ALP, AST, P, cholesterol, and Ca

⁵⁸ In addition, increases in BUN, serum TG, serum P, and serum K were observed.

gland weight, and hyperplasia and hyperkeratosis of the forestomach were observed in animals treated at ≥ 50 mg/kg; and reduced body weight gain, effects on red blood cell parameters (including decreased red blood cell count), increased serum Ca, decreased thymus weight, increased liver weight, mineralization of the glandular stomach, increased trabecular bone volume in the sternum and femur,⁵⁹ vascular mineralization in multiple organs (including the heart, kidneys, mammary gland, and mesenteric lymph node), effects on the kidneys (discolouration, cysts, surface disarrangement, dilatation of the renal tubules, exacerbation of chronic nephropathy, hyperplasia and mineralization of collecting tubular epithelial cells, acute inflammatory cell infiltration into the renal papilla), and increased frequency of mast cells in the mesenteric lymph node were observed in animals treated at 200 mg/kg. In the recovery groups, findings observed during the treatment period in animals treated at ≤ 50 mg/kg were found to be reversible, but animals treated at 200 mg/kg continued to show reduced body weight gain, increased water consumption, increased kidney weight, hyperplasia and hyperkeratosis of the forestomach, mineralization of the glandular stomach, increased trabecular bone volume in the sternum and femur, and effects on the kidneys (including dilatation of the renal tubules and exacerbation of chronic nephropathy). Based on the above, the NOAEL was determined to be 50 mg/kg/day.

3.(iii).A.(2).5 Six-month repeated oral dose toxicity study in rats (4.2.3.2.5)

Vehicle or dapagliflozin at doses of 5, 25, and 150 mg/kg was administered orally to male and female SD rats once daily for 6 months (with a reversibility assessment after a 3-month recovery period). Death occurred in animals treated at 150 mg/kg (17 of 60 animals [11 of 30 males, 6 of 30 females]). Increases in food and water consumption, urinary glucose, urine output, urinary Ca, urinary protein, BUN, and kidney weight, discolouration of the kidneys and adrenal gland, and increases in frequency and severity of hypertrophy and vacuolization of the zona glomerulosa of the adrenal cortex were observed in animals treated at ≥ 5 mg/kg; reduced body weight gain, effects on red blood cell parameters (including decreased red blood cell count), increased serum phosphorus (P), and increases in adrenal gland, liver, and heart weights were observed in animals treated at ≥ 25 mg/kg; increased platelet count, decreased urine osmolarity, increases in serum ALP, serum Ca, and spleen weight, vascular mineralization in multiple organs (including the heart, kidneys, mammary gland, mesentery, and aorta), increased trabecular bone volume in the sternum and femur,⁶⁰ effects on the kidneys (including dilatation of the renal tubules, hyperplasia of collecting tubular epithelial cells accompanied by mineralization, urothelial hyperplasia), mineralization of the tracheal mucosa, and enhanced extramedullary hematopoiesis in the spleen and liver were observed in animals treated at 150 mg/kg. In the recovery groups, findings observed during the treatment period in animals treated at ≤ 25 mg/kg were found to be reversible (excluding increased kidney weight), but animals treated at 150 mg/kg continued to show increases in food and water consumption, urine output, urinary glucose, and urinary protein, increased spleen weight, increased trabecular bone volume in the sternum and femur,⁶¹ vascular mineralization in the heart, and enhanced extramedullary hematopoiesis in the spleen. Based on the above, the NOAEL was determined to be 25 mg/kg/day.

3.(iii).A.(2).6 One-month repeated oral dose toxicity study in dogs (4.2.3.2.6)

Vehicle or dapagliflozin at doses of 5, 25, and 250 mg/kg was administered orally to male and female beagle dogs once daily for 1 month. Increases in water consumption, urinary glucose, and urine output, decreased urine osmolarity, and a trend toward decreased body weight were observed in animals treated at ≥ 5 mg/kg; and increased incidence of vomiting and loose stool,

⁵⁹ In animals treated with dapagliflozin, decreased blood 1,25-dihydroxyvitamin D, parathyroid hormone, and calcitonin levels were observed. In the 200 mg/kg group, increases in bone mineral density and bone mineral content in the distal femur and increased trabeculae volume in cancellous bone associated with mineralization based on histomorphometry of non-decalcified bone tissues.

⁶⁰ In animals treated with dapagliflozin, decreased urinary deoxypyridinoline and decreased 1,25-dihydroxyvitamin D (in males only) were observed. In the 150 mg/kg group, increases in bone mineral density, bone mineral content, and bone strength in the lumbar vertebra, and decreased bone formation rate in the femur were observed.

⁶¹ In female animals in the 150 mg/kg group, increases in bone mineral density, bone mineral content, and bone strength in the lumbar vertebra were observed.

increases in serum cholesterol and glucagon, decreased serum fructosamine, and increases in urinary Na and Ca were observed in animals treated at 250 mg/kg. Based on the above, the NOAEL was determined to be 25 mg/kg/day.

3.(iii).A.(2).7 Three-month repeated oral dose toxicity study in dogs (4.2.3.2.7)

Vehicle or dapagliflozin at doses of 5, 30, and 180 mg/kg was administered orally to male and female beagle dogs once daily for 3 months (with a reversibility assessment after a 1-month recovery period). Decreased body weight or a trend toward decreased body weight and increases in water consumption, urinary glucose, urine output, and urinary Na were observed in animals treated at \geq 5 mg/kg; increased incidence of vacuolization of proximal tubular epithelial cells and effects on red blood cell parameters (including decreased red blood cell count) were observed in animals treated at \geq 30 mg/kg; and increased incidence of vomiting, increases in urinary Ca and urinary protein, increased serum cholesterol, and prolongations of QT and QTc intervals (only in males at 13 weeks) were observed in animals treated at 180 mg/kg. In the recovery groups, increased urinary glucose was observed in animals treated at \geq 30 mg/kg, and increased serum cholesterol was observed in females treated at 180 mg/kg. Since the increased incidence and severity of vacuolization of the proximal renal tubules were found to be reversible and since no findings suggesting renal tissue damages were observed, the NOAEL was determined to be 30 mg/kg/day.

3.(iii).A.(2).8 Twelve-month repeated oral dose toxicity study in dogs (4.2.3.2.8)

Vehicle or dapagliflozin at doses of 5, 20, and 120 mg/kg was administered orally to male and female beagle dogs once daily for 12 months⁶² (with evaluations at 6 and 12 months [end of treatment] and a reversibility assessment after a 3-month recovery period after the end of treatment). A trend toward increases in food and water consumption, increases in urinary glucose, urine output, urinary Ca, Na, and P, and decreased urine osmolarity were observed in animals treated at \geq 5 mg/kg; increased watery/amorphous stool, and reduced body weight gain were observed in animals treated at \geq 20 mg/kg; and increases in urinary protein and serum cholesterol, increased adrenal gland weight, and decreased serum 1,25-dihydroxyvitamin D were observed in animals treated at 120 mg/kg. Inflammation of the renal pelvis and bladder (1 of 11 animals in the 5 mg/kg group, 1 of 11 animals in the 20 mg/kg group, 2 of 11 animals in the 120 mg/kg group), inflammation of the ureter (1 of 11 animals in the 5 mg/kg group, 1 of 11 animals in the 120 mg/kg group), and hyperplasia of transitional epithelium of the bladder and renal pelvis (1 of 11 animals in the 5 mg/kg group, 1 of 11 animals in the 120 mg/kg group) were observed in females in the dapagliflozin group, but the applicant explained that the inflammation of the renal pelvis and bladder was a change related to urinary tract infection, rather than a direct effect of dapagliflozin. In the recovery groups, findings observed during the treatment period were found to be reversible, and no effects on the bone tissue were observed in spite of the decreased urinary deoxypyridinoline observed in males in the 120 mg/kg group; therefore, the NOAEL was determined to be 120 mg/kg/day.

3.(iii).A.(3) Genotoxicity (4.2.3.3.1.1 to 4.2.3.3.1.4, 4.2.3.3.2.1 to 4.2.3.3.2.4)

A bacterial reverse mutation assay (Ames assay), a chromosomal aberration assay using CHO cells, a chromosomal aberration assay using rat peripheral blood lymphocytes, a 3-day oral micronucleus assay in rat, a 2-week oral micronucleus assay in rats, and a rat unscheduled DNA synthesis assay were conducted.

The Ames assay was negative, while in the chromosomal aberration assay using CHO cells, the number of cells with aberrant chromosome structure was increased dose-dependently by a 4-hour

⁶² Moribundity occurred in 1 of 11 females in the 5 mg/kg group (due to incorrect administration) and 1 of 11 males and 1 of 11 females in the 120 mg/kg group (due to tetracycline treated as labeling agent for histomorphometric measurement of non-decalcified bone tissues).

exposure to dapagliflozin at concentrations of ≥ 100 $\mu\text{g}/\text{mL}$ with metabolic activation by S9 (up to a 25% increase at 250 $\mu\text{g}/\text{mL}$). An additional testing was performed to evaluate clastogenicity of dapagliflozin and an amorphous form of dapagliflozin. The results for both samples showed an increase (43.9% and 46%, respectively) in the number of cells with aberrant chromosome structure at 300 $\mu\text{g}/\text{mL}$ under the same conditions.⁶³ Results of further evaluation of clastogenicity of dapagliflozin showed an increase (20%) in the number of cells with aberrant chromosome structure at 240 $\mu\text{g}/\text{mL}$.⁶⁴ The applicant explained that, based on the above, dapagliflozin is clastogenic with metabolic activation by S9. However, in the chromosomal aberration assay using rat peripheral blood lymphocytes,⁶⁵ no chromosomal aberrations were detected up to the maximum dose (200 mg/kg/day); the plasma exposure (AUC_{0-24h}) on Day 28 in animals treated at 200 mg/kg (1020 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males, 1210 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females) was ≥ 1400 -fold the plasma exposure³³ (AUC_{0-24h}) at the maximum recommended clinical dose. The rat micronucleus assay (3-day or 2-week repeated oral administration) was interpreted as negative since no increase in micronucleated polychromatic erythrocytes was observed up to the maximum tolerated dose (700 mg/kg/day) or the highest evaluable dose (250 mg/kg/day) of dapagliflozin. The rat unscheduled DNA synthesis assay (single oral administration) was interpreted as negative up to the maximum tolerated dose (700 mg/kg/day). Based on the above, dapagliflozin was not considered to have genotoxic potential *in vivo*.

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

Twenty-four months repeated oral dose studies of dapagliflozin in mice and rats were conducted. The plasma exposure (AUC) at a non-carcinogenic dose (40 mg/kg/day in male mice, 20 mg/kg/day in female mice, 10 mg/kg/day in rats) in mice and rats is respectively ≥ 40 -fold and ≥ 80 -fold the plasma exposure³³ (AUC_{0-24h}) at the maximum recommended clinical dose.

3.(iii).A.(4).1 Mouse carcinogenicity study (4.2.3.4.1.1)

Male and female CD-1 mice were treated orally with vehicle, distilled water, or dapagliflozin (at doses of 5, 15, and 40 mg/kg in males; and 2, 10, and 20 mg/kg in females) once daily for 24 months. Increased food consumption or a trend toward increased food consumption was observed in animals treated at ≥ 2 mg/kg after 6 months of treatment; decreased survival rate was observed in males treated at ≥ 15 mg/kg after 24 months of treatment; and increased body weight gain was observed in animals treated at 40 mg/kg at Week 101 of treatment (at the last observation). The applicant explained that no tumor development by dapagliflozin was observed and that the observed non-neoplastic lesions⁶⁶ (dilatation of the renal pelvis in males treated at ≥ 5 mg/kg, the bladder distention in animals treated at ≥ 15 mg/kg) were changes in response to the increased urine output. A slight increase in the incidence of hyperplasia of transitional epithelium of the bladder was observed in males treated with dapagliflozin (4 of 60 animals in the vehicle control group, 4 of 59 animals in the distilled water control group, 7 of 59 animals in the dapagliflozin 5 mg/kg group, 7 of 59 animals in the dapagliflozin 15 mg/kg group, and 6 of 60 animals in the dapagliflozin 40 mg/kg group).

3.(iii).A.(4).2 Rat carcinogenicity study (4.2.3.4.1.2)

Vehicle, distilled water, or dapagliflozin at doses of 0.5, 2, and 10 mg/kg was administered orally to male and female SD rats once daily for up to 90 weeks (in males)⁶⁷ or 105 weeks (in females). An increase in food consumption was observed in animals treated at ≥ 0.5 mg/kg after 6 months

⁶³ The cell proliferation inhibition rates with dapagliflozin and an amorphous form of dapagliflozin were 68% and 53%, respectively.

⁶⁴ The cell proliferation inhibition rate was 54%. Cytotoxicity was also evaluated by measuring decrease in intracellular ATP.

⁶⁵ Vehicle or dapagliflozin at doses of 25, 100, 150, and 200 mg/kg was administered orally to male and female SD rats (n = 10/group) once daily for 1 month to evaluate clastogenicity in peripheral lymphocytes. Animals in the positive control group (n = 5/group) received a single oral dose of cyclophosphamide (60 mg/kg).

⁶⁶ Findings are reported in the cases of increased incidence and/or severity as compared with the control group.

⁶⁷ Due to the low survival in the control (vehicle, distilled water) and 10 mg/kg groups, all male animals were necropsied at 89 to 91 weeks post-dose. The applicant explained that the main cause of the decreased survival in the 10 mg/kg group was exacerbation of chronic nephropathy.

of treatment; and reduced body weight gain or a trend toward reduced body weight gain was observed in animals treated at 10 mg/kg after 89 weeks (in males) and 101 weeks (in females) of treatment (at the last observation). No treatment-related tumor development was observed, and as non-neoplastic lesions,⁶⁶ vacuolization and hypertrophy of the zona glomerulosa of the adrenal gland in animals treated at ≥ 0.5 mg/kg, effects on the kidneys (cysts, vacuolization of the cortical tubular epithelium, atypical hyperplasia of the renal tubules,⁶⁸ exacerbation of chronic nephropathy) in male animals treated at ≥ 0.5 mg/kg, and increased trabecular bone volume in the sternum and femur in animals treated at 10 mg/kg were observed. The applicant explained that atypical hyperplasia of the cortical tubules is a reaction associated with the exacerbation of chronic nephropathy rather than a change suggestive of precancerous lesions because no renal cancer was observed in this study (Hard GC, et al. *Toxicol Pathol.* 2004;32:171-80). In addition, hyperplasia of transitional epithelium of the bladder was observed (males, 0 of 70 animals in the vehicle control group, 2 of 70 animals in the distilled water control group, and 0 of 70 animals in the dapagliflozin 0.5 mg/kg group, 3 of 70 animals in the dapagliflozin 2 mg/kg group, and 0 of 70 animals in the dapagliflozin 10 mg/kg group; females, 0 of 70 animals in the vehicle control group, 1 of 70 animals in the distilled water control group, and 2 of 70 animals in the dapagliflozin 0.5 mg/kg group, 0 of 70 animals in the dapagliflozin 2 mg/kg group, and 3 of 70 animals in the dapagliflozin 10 mg/kg group).

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

A study of fertility and early embryonic development to implantation (rats), embryo-fetal development studies (rats, rabbits), and a study on pre- and postnatal development, including maternal function (rats) were conducted. In the study of fertility and early embryonic development to implantation, toxic effects on male reproductive organs were observed (including decreases in epididymis and vesicular gland weights, a decrease in sperm counts, and morphologic abnormalities). In the embryo-fetal development studies in rats, incomplete ossification including bifid thoracic vertebra and sternebrae, and cardiovascular anomaly were observed. In the study on pre- and postnatal development including maternal function in rats, decreased body weight and retardation in preputial separation were observed in F₁ pups. The plasma AUCs in rats and rabbits at the NOAEL (75 and 180 mg/kg/day, respectively) observed in the embryo-fetal development studies are ≥ 900 -fold and approximately 760-fold, respectively, the plasma exposure³³ (AUC_{0-24h}) at the maximum recommended clinical dose.

3.(iii).A.(5).1 Study of fertility and early embryonic development to implantation in rats (4.2.3.5.1.1)

Vehicle or dapagliflozin at doses of 15, 75, and 300/210⁶⁹ mg/kg was administered orally to male SD rats once daily from 2 weeks prior to mating until the day of necropsy (for at least 43 days), and vehicle or dapagliflozin at doses of 3, 15, and 75 mg/kg was administered orally to female SD rats once daily from 2 weeks prior to mating until Gestation day 7. Death (2 of 50 animals [2 of 25 males]) and moribundity (9 of 50 animals [8 of 25 males, 1 of 25 females]) occurred in the high dose group.⁷⁰ Increased food consumption,⁷¹ reduced body weight gain (males, throughout the treatment period; females, during premating treatment period), decreased prostate weight and chromorhinorrhea were observed in animals treated at ≥ 15 mg/kg; red attached substances around the mouth, salivation, and rales were observed in animals treated at ≥ 75 mg/kg; and worsening of clinical conditions (including dehydration, hunched position, coarse fur, abdominal distension, decreased locomotor activity, and tremor), decreased weights or a trend toward decreased weights of testis, epididymis and vesicular gland, decreased sperm motility, decreased sperm counts in the

⁶⁸ In 27 of 70 animals in the vehicle control group, 18 of 70 animals in the distilled water control group; and 44 of 70 animals, 42 of 70 animals, and 45 of 70 animals in the dapagliflozin 0.5, 2, and 10 mg/kg groups, respectively.

⁶⁹ Excessive toxicity was observed during the first 4 days in males treated at 300 mg/kg/day, and the dose was changed to 210 mg/kg/day from Day 5.

⁷⁰ Death occurred in 1 of 10 males in the high dose group for toxicokinetic analysis on Day 14.

⁷¹ Decreased food consumption was observed in males in the high dose group up to Day 8.

cauda epididymis, and increased number of sperms with morphologic abnormalities (separation or absence of the sperm head) were observed in males in the high dose group. The applicant explained that, given the absence of reports of infertility both in patients with familial renal glycosuria, who have mutations in SGLT2 gene and in SGLT2-KO mice (Vallon V, et al. *J Am Soc Nephrol.* 2011;22:104-12), and given the fertilization ability observed in SGLT1-KO mice (Gorboulev V, et al. *Diabetes.* 2012;61:187-96), the effects on spermatogenesis observed in males in the high dose group are changes associated with the worsening of clinical conditions rather than direct effects of dapagliflozin. Based on the above, the NOAEL for clinical conditions was determined to be 75 mg/kg/day in males and 15 mg/kg/day in females, and the NOAEL for reproductive performance and early embryonic development was determined to be 75 mg/kg/day in both males and females.

3.(iii).A.(5).2) Study on embryo-fetal development in rats (4.2.3.5.2.1)

Vehicle or dapagliflozin at doses of 37.5, 75, 150, and 300 mg/kg was administered orally to pregnant SD rats once daily on Gestation day 6 to Gestation day 15. Death (3 of 25 animals [1 of 25 animals on Gestation day 10, 2 of 25 animals on Gestation day 16]) and moribundity (1 of 25 animals on Gestation day 15) occurred in the 300 mg/kg group; these animals showed worsening of clinical conditions including pallor of the extremities, decreased locomotor activity, and labored respiration.⁷² In maternal animals, reduced body weight gain during the treatment period was observed in the ≥ 37.5 mg/kg groups; decreased food consumption, loose stool, chromorhinorrhea, dehydration, salivation, stained fur, coarse fur, exudate around the mouth, and rales were observed in the ≥ 150 mg/kg groups; and local alopecia was observed in the 300 mg/kg group. In embryos and fetuses, decreased fetal weight, skeletal anomaly,⁷³ and vascular anomaly⁷⁴ were observed in the ≥ 150 mg/kg groups; and a decrease in the number of live fetuses, an increase in early and late resorptions, an increase in postimplantation loss, and an increase in the percentage of male fetuses were observed in the 300 mg/kg group. The applicant explained that the effects on embryos and fetuses were not direct effects of dapagliflozin because these effects had occurred at the maternally toxic doses. Based on the above, the NOAELs for maternal general and reproductive toxicity and embryo-fetal development were all determined to be 75 mg/kg/day.

3.(iii).A.(5).3) Study on embryo-fetal development in rabbits (dose-finding study, 4.2.3.5.2.5)

Vehicle or dapagliflozin at doses of 37.5, 75, 150, and 300 mg/kg was administered orally to pregnant NZW rabbits once daily from Gestation day 7 to Gestation day 19. Reduced body weight gain was observed in the early phase of dosing in the ≥ 37.5 mg/kg groups; decreased food consumption was observed in the ≥ 150 mg/kg groups (only for 1 week after the start of administration); and increased late resorptions were observed in the 300 mg/kg group.

3.(iii).A.(5).4) Study on embryo-fetal development in rabbits (4.2.3.5.2.4)

Vehicle or dapagliflozin at doses of 20, 60, and 180 mg/kg was administered orally to pregnant NZW rabbits once daily from Gestation day 7 to Gestation day 19. In maternal animals, reduced body weight gain from Gestation day 7 to Gestation day 20 was observed in the dapagliflozin groups; and decreased fecal volume, loose stool, and stained fur were observed in the 180 mg/kg group. No effects were observed in embryos and fetuses. The NOAEL for maternal animals, embryos, and fetuses was determined to be 180 mg/kg/day.

⁷² Moribundity occurred in 2 of 12 animals in the 300 mg/kg group for toxicokinetic analysis.

⁷³ Expansion of the frontonasal suture, and incomplete ossification including bifid thoracic vertebra and sternebrae were observed in the ≥ 150 mg/kg groups; and fusion of the thoracic spine, lumbar spine, and fused ribs, overlap of the sternum, etc., were observed in the 300 mg/kg group.

⁷⁴ Cardiovascular anomaly was observed in 1 of 25 animals in the 150 mg/kg group and 3 of 25 animals in the 300 mg/kg group; and vascular malformation in the subclavian vessels and/or umbilical area was observed in 3 of 25 animals in the 300 mg/kg group (including the 2 animals with cardiovascular anomaly).

3.(iii).A.(5).5 Rat study on pre- and postnatal development, including maternal function (4.2.3.5.3.1)

Vehicle or dapagliflozin at doses of 1, 15, and 75 mg/kg was administered orally to pregnant SD rats once daily from Gestation day 6 through Lactation days 20 to 22. In maternal animals, increased food consumption was observed in the ≥ 1 mg/kg groups; and reduced (during pregnancy) or increased (during lactation) body weight gain, salivation, and stained fur around the mouth were observed in the 75 mg/kg group. In the offspring (F_1), reduced body weight gain (during lactation) and a trend toward decreased body weight (after weaning) were observed in the ≥ 15 mg/kg groups; and decreased infant birth weight, retardation in preputial separation, increased incidence (males) and severity (females) of dilatation of the renal pelvis were observed in the 75 mg/kg group. The applicant explained that, given the probable exposure of F_1 offspring to dapagliflozin via milk due to the observed excretion of dapagliflozin in milk⁷⁵ and the kidney development in rats requiring a period of up to Postnatal day 11 (Zoetis T. et al. *Birth Defects Res (part B)*. 2003;68:111-120), dapagliflozin may affect the developing kidneys. No effects on the reproductive function of the F_1 generation or abnormalities in the F_2 generation were observed. Based on the above, the NOAEL was determined to be 75 mg/kg/day for maternal clinical conditions and reproductive function of the F_1 generation and 1 mg/kg/day for F_1 clinical conditions.

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

3.(iii).A.(6).1 Mechanistic studies of toxicity

Since effects on Ca homeostasis (including increases in serum Ca and urinary Ca excretion, and vascular mineralization in various organs) and increased urinary protein were seen in rats treated with dapagliflozin, and since bladder cancer was seen in clinical studies, mechanistic studies on these events were conducted.

(a) Ten-day repeated oral dose toxicity study evaluating effects on Ca and bone in female rats (Reference data, 4.2.3.7.3.1)

Vehicle or dapagliflozin 250 mg/kg was orally administered to female SD rats once daily for 10 days (animals in each group were fed with the normal diet or glucose-free diet⁷⁶), and effects of dapagliflozin on Ca homeostasis were evaluated in relation to SGLT1 inhibition.⁷⁷ The results showed no effects of diet type on the plasma exposure to dapagliflozin,⁷⁸ and pharmacological activity (urinary glucose excretion) was similar regardless of diet type.⁷⁹ Death occurred in an animal receiving dapagliflozin in the normal diet group (1 of 10 animals). Increased incidences of loose stool and coarse fur, etc., gaseous and watery cecal contents (9 of 9 animals), decreased pH of cecal contents (1 of 9 animals), mineralization of the glandular stomach mucosa (6 of 9 animals), regressive changes in the renal tubules associated with regenerative hyperplasia (9 of 9 animals), increases in incidence and severity of atrophy of white adipose tissue in abdominal organs, and increases in incidence and severity of single cell necrosis of the cecal mucosa were observed. In addition, urinary Ca, P, and protein levels increased⁸⁰ and were 10.8, 4.2, and 6.9 times higher, respectively, in the normal diet group and 2.3, 1.7, and 1 times higher, respectively, in the glucose-free diet group than those in the control group. The applicant explained that the evident increase in urinary Ca and the mineralization of the glandular stomach mucosa observed

⁷⁵ Concentrations in milk at 2 hours post-dose on Lactation day 10 in the 1, 15, and 75 mg/kg groups were 0.290, 2.986, and 10.130 $\mu\text{g}/\text{mL}$, respectively.

⁷⁶ Diet prepared by replacing glucose with fructose

⁷⁷ Because fructose is not a substrate of SGLT1, it has been considered that the impact of SGLT1 inhibition could be minimized by feeding the diet prepared by replacing glucose with fructose.

⁷⁸ Plasma concentrations at 2 and 4 hours post-dose on Day 10 were 67.1 and 24.4 $\mu\text{g}/\text{mL}$, respectively, in the normal diet group, and 99.7 and 31.6 $\mu\text{g}/\text{mL}$, respectively, in the glucose-free diet group.

⁷⁹ Glucose excretion in 18-hour urine in the normal diet group was approximately 1963 times, and that in the glucose-free diet group was approximately 3820 times higher than that in the control group. The glucose/creatinine ratio obtained from urinalysis on fresh urine in the normal diet group was approximately 1963 times, and that in the glucose-free diet group was approximately 1103 times higher than that in the control group.

⁸⁰ The ratio of mean values (dapagliflozin/control)

in the normal diet group compared with the glucose-free diet group indicated that dapagliflozin inhibits SGLT1 in the rat intestine and induces a change in Ca homeostasis.

(b) One-month repeated oral dose toxicity study on the mechanism of proteinuria in female rats (Reference data, 4.2.3.7.3.5)

Vehicle or dapagliflozin 50 mg/kg was administered orally to female SD rats once daily for 1 month. Increases in serum ALT, serum TG, and BUN, and increases in AST and ALT mRNA expressions in the liver (1.5 and 3.0 times higher than in the control group, respectively) were observed on Day 21 in animals treated with dapagliflozin. The applicant explained that the toxicological significance of the decreased AST and ALT levels was limited because alterations in gluconeogenesis and nutritional status were known to affect ALT and AST gene expressions (Kobayashi A, et al. *J Toxicol Sci*. 2001;36:325-37) and because no histopathological findings in hepatocytes were noted in the previous repeated oral dose toxicity studies. In addition, urinalysis on Days 14 and 28 revealed increases in glucose, urine output, urinary specific gravity (on Day 14 only), osmolarity (on Day 14 only), total protein, Ca, P, Na, and total N-acetyl- β -D-glucosaminidase (NAG). The applicant explained that the increases in urinary excretion of NAG and total protein observed in this study were secondary effects related to the pharmacological activity of dapagliflozin for the following reasons: (i) the degree of these increases was mild;⁸¹ (ii) no alterations of the kidneys were found on necropsy or histopathological examination; (iii) the similar urinary protein profiles⁸² between the control and dapagliflozin groups and the absence of increased urinary albumin in animals in the dapagliflozin group suggest that dapagliflozin has no effect on the glomerular function; and (iv) diuretics has been reported to induce increases in urinary excretion of total protein and NAG without apparent nephrotoxicity in rats.⁸³

(c) Study in SGLT2-knockout mice on the potential for inducing human bladder cancer (Reference data, 4.2.3.7.3.6)

The effects on morphology of the urinary tract and on renal function were evaluated in male and female SGLT2-KO mice compared with those in wild-type mice. SGLT2-KO mice and wild-type mice were maintained under ad libitum food (18% protein diet) and water conditions until 15 months of age. The results showed no difference in survival to 15 months of age between SGLT2-KO mice (86% [31 of 36 animals (20 of 23 males, 11 of 13 females)]) and wild-type mice (85% [28 of 33 animals (15 of 16 males, 13 of 17 females)]). No urinary glucose was noted in wild-type mice, while a marked urinary glucose excretion (approximately 2000 mg/dL) was noted in SGLT2-KO mice. The applicant explained that a prolonged urinary glucose excretion was unlikely to induce renal impairment or bladder tumors because no proliferative changes were seen in the kidneys or bladder of SGLT2-KO mice.

3.(iii).A.(6).2 Studies on metabolites

Although toxicity of metabolites of dapagliflozin was not studied, toxicokinetic studies were conducted to measure the exposures of dapagliflozin 3-O-glucuronide (a major human metabolite) in rats and dogs.

(a) Rat single-dose toxicokinetic study (4.2.3.7.5.1)

A single dose of dapagliflozin 150 mg/kg was administered to male and female SD rats. The plasma exposure (AUC_{0-24h}) to dapagliflozin was 992 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males and 1250 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females and the plasma exposure (AUC_{0-24h}) to dapagliflozin 3-O-glucuronide was 3.53 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males and 4.44 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females (both values are equivalent to 0.36% of the exposure to

⁸¹ NAG was increased by 2.4 times, and total protein was increased by 1.6 to 2 times compared with the control group.

⁸² The urinary protein profiles (bands corresponding to albumin and a molecule with molecular weight of <30 kDa were detected by polyacrylamide-gel electrophoresis of urinary proteins with coomassie blue staining) after 2 and 4 weeks of treatment were similar to those observed in the control group.

⁸³ Kutina AV, et al. *Bull Exp Bio Med*. 2008;146:613-6; Lina BAR, et.al. *Regul Toxicol Pharmacol*. 1996;24:264-79.

unchanged dapagliflozin). Based on the plasma exposure (AUC_{0-24h}) to dapagliflozin at the NOAEL observed on Day 180 in the 6-month repeated oral dose toxicity study in rats (161 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males, 314 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females), the plasma exposure (AUC_{0-24h}) to dapagliflozin 3-*O*-glucuronide was estimated to be 0.58 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males and 1.13 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females, which are equivalent to approximately 0.37- and 0.72-fold, respectively, the plasma exposure⁸⁴ (AUC_{0-24h}) at the maximum recommended clinical dose.

(b) Dog single-dose toxicokinetic study (4.2.3.7.5.2)

A single dose of dapagliflozin 120 mg/kg was administered to male and female beagle dogs. Vomiting was observed in 2 of 4 males and 2 of 4 females within 1 hour post-dose. The plasma exposure (AUC_{0-24h}) to dapagliflozin was 1280 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males and 1710 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females, and the plasma exposure (AUC_{0-24h}) to dapagliflozin 3-*O*-glucuronide was 55.9 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males and 106 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females (equivalent to 4.4% and 6.2% of the exposure to unchanged dapagliflozin, respectively). Based on the plasma exposure (AUC_{0-24h}) to dapagliflozin at the NOAEL observed on Day 363 in the 12-month repeated oral dose toxicity study in dogs (1520 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males, 1540 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females), the plasma exposure (AUC_{0-24h}) to dapagliflozin 3-*O*-glucuronide was estimated to be 76 $\mu\text{g}\cdot\text{h}/\text{mL}$ in males and 77 $\mu\text{g}\cdot\text{h}/\text{mL}$ in females, which are equivalent to approximately 48-fold the plasma exposure³³ (AUC_{0-24h}) at the maximum recommended clinical dose.

3.(iii).A.(6).3 Study in juvenile animals (Reference data, 4.2.3.7.7.4)

Vehicle or dapagliflozin at doses of 1, 15, and 75 mg/kg was administered orally to male and female juvenile SD rats once daily on postnatal days 21 to 90 (with a reversibility assessment after a 1-month recovery period).⁸⁵ Increased food consumption, increases or a trend toward increases in urinary glucose, urine output, urinary Ca, BUN, serum ALT, and serum AST, increased kidney weight, vacuolization of the adrenal glomerulosa, and effects on the kidneys (hypertrophy, increased incidence of dilatation/mineralization of the renal pelvis and tubules) were observed in the ≥ 1 mg/kg groups; increases in urinary Na and P and a decrease in serum glucose were observed in the ≥ 15 mg/kg groups; and decreased serum protein and increased trabecular bone volume in the sternum were observed in the 75 mg/kg group. As changes found only in juvenile animals (not in adult animals), a trend toward decreases in serum cholesterol and TG in males in the dapagliflozin group; decreases in white blood cell count, lymphocyte count, and serum Ca, a trend toward decreases in serum chloride and total carbon dioxide levels, increased incidences of erosion/ulcer and haemorrhage of the gastric mucosa, and decreased pancreatic Zymogen granules in the ≥ 15 mg/kg groups; and a trend toward decreased body weight (decreased by approximately 10% compared with the control group) and decreased length of head to hip in the 75 mg/kg group were observed. In the recovery groups, the vacuolization of the adrenal glomerulosa (only in females) and effects on the kidneys (including dilatation of the renal pelvis and tubules) were found to be irreversible, and animals in the ≥ 15 mg/kg groups continued to show increased urine output. Regarding the observed trend toward higher severity compared with adult animals (e.g., erosion/ulcer of the gastric mucosa, changes in the findings of clinical chemistry), the applicant explained that this was due to insufficient capability of juvenile animals to produce compensatory metabolic responses against pharmacological activity of drugs, and that especially the effects on the pancreas and stomach were changes typical of stress related to the long-term energy deprivation or hypoglycaemia.⁸⁶ In addition, the applicant explained that the irreversibility of the effects on the kidneys (including dilatation of the renal tubules and pelvis)

⁸⁴ The plasma AUC_τ (1581 $\text{ng}\cdot\text{h}/\text{mL}$) of dapagliflozin 3-*O*-glucuronide on Day 14 observed in a clinical pharmacology study (Study MB102025, 5.3.3.2.1), in which dapagliflozin was orally administered once daily for 14 days in Japanese patients with type 2 diabetes mellitus at the maximum recommended clinical dose.

⁸⁵ Mating performance and fertility were evaluated after the end of treatment (11 of 21 animals/group).

⁸⁶ Macdonald A, et.al. *Am J Dig Dis.* 1997;22:909-14, Kinoshita M, et.al. *Aliment Pharmacol Ther.* 2000;14:359-66

was due to inadequate adaptability of the developing rat kidneys⁸⁷ to increased urine output. Furthermore, the applicant explained that, given the increases in incidence (males) and severity (females) of dilatation of the renal pelvis observed in offspring (F_1) in the study on pre- and postnatal development including maternal function in rats (4.2.3.5.3.1), the risks from exposure of the developing human kidneys⁸⁷ to dapagliflozin could not be ruled out, and therefore, a caution statement would be included in the package insert regarding the impacts during human pregnancy corresponding to the studied period in juvenile animals. Based on the above, only the NOAEL for reproductive function was determined (75 mg/kg/day).

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

PMDA accepted the applicant's response from a toxicological point of view based on reviews of (1) to (3) described below. However, nephrotoxicity on humans will be additionally reviewed in the clinical sections [see "4.(iii).B.(3).7 Renal disorder"]. Given the data from the study in SGLT2-KO mice on the potential for inducing human bladder cancer (Reference data, 4.2.3.7.3.6), the applicant's explanation that a prolonged urinary glucose excretion is unlikely to induce renal or bladder tumors is acceptable. However, the relationship between dapagliflozin and human bladder cancer as well as the safety in humans will be discussed in the clinical sections [see "4.(iii).B.(3).11 Malignant tumor risk"].

3.(iii).B.(1) Nephrotoxicity

Regarding the effects on the kidneys of dapagliflozin observed in rats, PMDA asked the applicant to explain the mechanism and safety in humans.

The applicant responded as follows:

The effects on the kidneys observed in rats are considered to be due to mineralization, which is attributed to the increased serum Ca caused by the enhanced Ca absorption due to SGLT1 inhibition. This is supported by the facts that regressive changes in the renal tubules associated with regenerative hyperplasia were seen in the normal diet (containing glucose) group but not in the glucose-free diet group in the 10-day repeated oral dose toxicity study on effects on Ca and bone in female rats (4.2.3.7.3.1), and that nephrocalcinosis and proximal renal tubular dysfunction have been reported to occur in humans with glucose-galactose malabsorption syndrome who have genetic deficiencies in SGLT1.⁸⁸ The hyperplasia observed in the renal tubules, collecting tubules, and urothelium is considered to be a reactive change associated with regeneration/repair after tissue damage. These findings were only seen at exposures that are ≥ 1600 times higher than the plasma exposure³³ at the maximum recommended clinical dose. In addition, exacerbation of chronic nephropathy and atypical hyperplasia were observed in male rats in the rat carcinogenicity study, but these findings are perceived as inappropriate for extrapolation to humans, because chronic nephropathy is an age-related spontaneous pathological change that is normally observed in SD rats but humans do not have its equivalent.⁸⁹ Based on the above, safety concerns in humans are considered to be limited regarding the effects of dapagliflozin on the kidneys.

The dapagliflozin-related change in Ca homeostasis is considered to be due to (a) glucose malabsorption via inhibition of SGLT1 activity in the intestine, (b) enhanced bacterial fermentation and decreased intestinal pH associated with increased amount of intestinal glucose, and (c) increased Ca absorption associated with decreased gastrointestinal pH. SGLT1 is a major glucose co-transporter in the intestine, and humans lacking SGLT1 activity suffer from a disease called glucose-galactose malabsorption syndrome; symptoms consistent with those seen in these patients (e.g., loose stool, abdominal distension due to gas evolution) have been observed also in

⁸⁷ It has been known that renal development in humans occurs at around week 18 to 32 of pregnancy and completes while in the uterus at week 35 of pregnancy in contrast to rats, in which renal development continues until approximately 6 weeks of age (Zoetis T, et al. *Birth Defects Res (part B)*. 2003;68:111-120)

⁸⁸ Soylu OB, et al. *Eur J Pediatr*. 2008;167:1395-8

⁸⁹ Hard GC, et.al. *Toxicol Pathol*. 2004;32:171-80, Hard GC, et.al. *Toxicol Sci*. 2013;132:268-75

mice, rats, and dogs treated with dapagliflozin at a high dose. Expression levels of SGLT1 in the gastrointestinal tract and duration of SGLT1 inhibition in each animal species were not evaluated. However, the selectivity of dapagliflozin to SGLT2 versus SGLT1 in humans is higher than that in rats (approximately 200 times selective in rats, 1200 times in humans) (4.2.1.1.2), and the estimated unbound plasma C_{max} of dapagliflozin at the NOAEL in the 6-month repeated oral dose toxicity study in rats (4.2.3.2.5) and 12-month repeated oral dose toxicity study in dogs (4.2.3.2.8) (approximately 8 and 30 µM, respectively) were above the IC₅₀ value for SGLT1 inhibition (0.620 µM in rats, 0.698 µM in dogs). Thus, it is considered that the toxicity due to SGLT1 inhibition has been adequately examined. In addition, increased adverse events of diarrhoea were not observed in clinical studies. Therefore, SGLT1 inhibition is unlikely to occur at the maximum recommended clinical dose, and safety concerns in humans are considered to be limited. Furthermore, the 12-month repeated oral dose toxicity study in dogs (4.2.3.2.8) evaluated a dose at which exposure is >2000 times higher than that at the maximum recommended clinical dose,³³ but showed no toxicity. Increased trabecular bone volume and mineralization of blood vessels etc. in various organs were observed in rats. However, differences between species exist in the selectivity of dapagliflozin to SGLT2 versus SGLT1 as well as in bone physiology; the above findings were observed only when the exposure exceeded 1300-fold that at the maximum recommended clinical dose;³³ no clinically significant differences in percent change in bone mineral density were seen in the dapagliflozin group compared with the control group in Foreign Study D1690C00012; and no events suggestive of effects on the intestine, trachea, and heart were seen in this clinical study. Given the above, safety concerns in humans due to SGLT1 inhibition are considered to be limited.

PMDA asked the applicant to explain the potential concerns about increase of nephrotoxicity risk associated with the urinary glucose excretion promoting activity of dapagliflozin.

The applicant responded as follows:

The study in SGLT2-KO mice on the potential for inducing human bladder cancer (4.2.3.7.3.6) evaluated the effects of lifelong exposure to high concentration urinary glucose on the kidneys, but showed neither histopathological findings in the kidneys nor findings suggestive of renal impairment. Therefore, concerns about increase of nephrotoxicity risk associated with the urinary glucose excretion promoting activity are limited.

PMDA accepted the applicant's response that safety concerns in humans related to SGLT1 inhibition are limited based on the following facts: dapagliflozin is 1200 times more selective to SGLT2 than to SGLT1 in humans; across toxicity studies which evaluated an exposure that is ≥1000-fold higher than that at the maximum recommended clinical dose,³³ the only common findings related to SGLT1 inhibition were the effects on the gastrointestinal system (abdominal distension, diarrhoea, loose stool); and adverse events such as diarrhoea did not increase in patients who received dapagliflozin in clinical studies.

3.(iii).B.(2) Hyperplasia of transitional epithelium

PMDA asked the applicant to explain potential relationship between dapagliflozin and hyperplasia of transitional epithelium of the renal pelvis and/or bladder observed in the 12-month repeated oral dose toxicity study in dogs and carcinogenicity studies in rats and mice.

The applicant responded as follows:

In dogs, the hyperplasia of transitional epithelium of the renal pelvis and bladder was observed in 1 female each in the 5 mg/kg/day and 120 mg/kg/day groups. These are considered to be reactive hyperplasia associated with ascending urinary tract infection because all these animals also showed inflammation of the renal pelvis and bladder. In addition to these 2 females with hyperplasia, inflammation of the renal pelvis and bladder occurred in 1 female each in the

20 mg/kg/day and 120 mg/kg/day groups; overall, only 4 females out of a total of 66 male and female beagle dogs treated with dapagliflozin showed such inflammation. In addition, given the facts that the 2 animals with hyperplasia were necropsied at the end of 6-month treatment while no animals necropsied at the end of 12-month treatment showed hyperplasia and that the incidence/severity of findings related to urinary tract infection did not show a dose- or time-dependent increase, the urinary tract infection and hyperplasia of transitional epithelium in dogs are not considered to be dapagliflozin-related changes. In the rat carcinogenicity study, hyperplasia of transitional epithelium was observed in the bladder (males, 0 of 70 animals in the vehicle control group, 2 of 68 animals in the distilled water control group, 0 of 70 animals in the dapagliflozin 0.5 mg/kg group, 3 of 67 animals in the dapagliflozin 2 mg/kg group, 0 of 70 animals in the 10 mg/kg group; females, 0 of 70 animals in the vehicle control group, 1 of 69 animals in the distilled water control group, 2 of 68 animals in the dapagliflozin 0.5 mg/kg group, 0 of 70 animals in the dapagliflozin 2 mg/kg group, 3 of 67 animals in the 10 mg/kg group), but the difference was not statistically significant and 3 of 5 males and 4 of 6 females with hyperplasia showed inflammation of the bladder, thus, the finding is considered unrelated to dapagliflozin. In the mouse carcinogenicity study, hyperplasia of transitional epithelium was observed in the bladder (males, 4 of 56 animals in the vehicle control group, 4 of 56 animals in the distilled water control group, 7 of 53 animals in the dapagliflozin 5 mg/kg group, 7 of 53 animals in the dapagliflozin 15 mg/kg group, 6 of 54 animals in the dapagliflozin 40 mg/kg group) without statistical significance as compared with the control group. Most of the animals with hyperplasia of transitional epithelium in the bladder (4 of 4 animals in the vehicle control group, 3 of 4 animals in the distilled water control group, 7 of 7 animals in the dapagliflozin 5 mg/kg group, 5 of 7 animals in the dapagliflozin 15 mg/kg group, 6 of 6 animals in the 40 mg/kg group) exhibited urogenital syndrome,⁹⁰ which is commonly observed in male CD-1 mice, suggesting a potential relationship with progression of mouse urogenital syndrome. Based on the above, the hyperplasia in the bladder was not directly induced by dapagliflozin but is a change related to mouse urogenital syndrome.

PMDA considers as follows:

Since the animals in which urogenital syndrome was diagnosed included those considered as such based solely on findings of bladder dilatation and inflammation of the prostate and/or vesicular gland, which have an unclear relationship with hyperplasia of transitional epithelium of the bladder. Therefore, relationship between urologic syndrome and the hyperplasia of transitional epithelium is unknown. However, PMDA accepted the applicant's response that the urothelial hyperplasia is unlikely to be caused by dapagliflozin based on the facts that findings suggestive of the relation with hyperplasia of transitional epithelium (e.g., inflammation, calculus, mineralization) were observed in 1 of 4 animals (25%) in the vehicle control group, 2 of 3 animals (67%) in the distilled water control group, and 5 of 7 animals (71%) in the dapagliflozin 5 mg/kg group, 5 of 5 animals (100%) in the dapagliflozin 15 mg/kg group, and 2 of 6 animals (33%) in the 40 mg/kg group, and that, among female mice, the hyperplasia of transitional epithelium was seen in only 1 animal in the distilled water control group.

3.(iii).B.(3) Clastogenicity of metabolites

Given the increased frequency of cells with aberrant chromosome structure under metabolic activation by S9 observed in the chromosomal aberration assay using CHO cells, PMDA asked the applicant to explain the safety in humans.

⁹⁰ Background lesions commonly observed in male mice, consisting of urogenital inflammation or bladder distension, known to lead to partial or complete urogenital disturbance (Gaillard ET. *Pathology of the mouse-reference and atlas*, 1st ed. Ed. by Maronpot RR. Cache River Press. 1999;250-3)

The applicant responded as follows:

In the *in vitro* chromosomal aberration assay, clastogenic potential was seen only under metabolic activation by rat S9 (at concentrations of ≥ 100 $\mu\text{g/mL}$). Thus, the clastogenic potential was attributed to metabolite(s). Evaluation of *in vitro* metabolism of dapagliflozin using Aroclor-induced rat S9 fraction revealed production of oxidative metabolites (desethyl dapagliflozin [m8], benzylic hydroxy-dapagliflozin [m12]) (4.2.2.2.1). The estimated plasma exposures⁹¹ (AUC) of desethyl dapagliflozin⁹² and benzylic hydroxy-dapagliflozin at a dose determined as non-carcinogenic in the carcinogenicity studies (40 mg/kg/day in mice, 10 mg/kg/day in rats) were approximately 10,241 and 12,241 ng·h/mL, respectively, in mice; and 658 and 1972 ng·h/mL, respectively, in rats. The estimated human plasma exposures (AUC) to desethyl dapagliflozin and benzylic hydroxy-dapagliflozin at the maximum recommended clinical dose were approximately 5 and 40 ng·h/mL, respectively, resulting in the exposure ratios for each metabolite (desethyl dapagliflozin and benzylic hydroxy-dapagliflozin) of approximately 2200 and 300, respectively, for the mice; or 140 and 49, respectively, for the rats. The estimated urinary concentrations⁹³ of these metabolites (desethyl dapagliflozin⁹⁴ and benzylic hydroxy-dapagliflozin) were calculated to be approximately 40 and 12 $\mu\text{g/mL}$, respectively, in mice; 5.3 and 2.3 $\mu\text{g/mL}$, respectively, in rats; and 0.011 and 0.038 $\mu\text{g/mL}$, respectively, in humans. Desethyl dapagliflozin coeluted with other metabolites both in rats and mice, and therefore it is difficult to adequately estimate its urinary concentrations, but these are considered to present at substantially higher concentrations compared with those in humans. Given the facts that rat micronucleus assay (4.2.3.3.2.2, 4.2.3.3.2.3) was negative and that no tumorigenic potential was observed in the carcinogenicity studies of dapagliflozin in mice and rats, the clastogenic potential of this metabolite is unlikely to become relevant in humans.

PMDA accepted the applicant's response that the metabolites, that are responsible for the clastogenic potential (desethyl dapagliflozin, benzylic hydroxy-dapagliflozin), are unlikely to become relevant in humans, based on the fact that the estimated plasma exposure to desethyl dapagliflozin in rats (calculated as desethyl dapagliflozin alone) was approximately 140-fold that at the maximum recommended clinical dose although exposure to desethyl dapagliflozin in mice (calculated as the total amount with dapagliflozin carboxylic acid [m7] and hydroxy-dapagliflozin-2 [m9]) represents an high estimation in terms of exposure ratios.

4. Clinical data

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

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

Five formulations (capsules, uncoated tablets, 2 kinds of film-coated tablets,⁹⁵ proposed commercial formulation) were used in the clinical development of dapagliflozin. Details of formulations used in clinical studies are shown in Table 4. The proposed commercial formulation is film-coated tablets, which has been demonstrated to be bioequivalent with the film-coated tablets used in the phase III studies based on dissolution testing complying with "Guideline for Bioequivalence Studies for Formulation Changes of Oral Solid Dosage Forms" (PMSB/ELD Notification No. 67 dated February 14, 2000, partially revised by PFSB/ELD Notification No. 1124004 dated November 24, 2006).

⁹¹ Plasma exposures in rats, mice, and humans were estimated based on the results from a pharmacokinetic study (4.2.2.4.2) of ^{14}C -dapagliflozin.

⁹² Was calculated as the total amount of the following 3 compounds because the former 2 compounds coeluted with desethyl dapagliflozin in mice: dapagliflozin carboxylic acid (m7), hydroxy-dapagliflozin-2 (m9), and desethyl dapagliflozin.

⁹³ Were calculated based on the relative percent distribution of metabolites in urine samples determined in a pharmacokinetic study (4.2.2.4.2) of ^{14}C -dapagliflozin. For mice, 12-hour urine was used, and for rats and humans, 24-hour urine was used.

⁹⁴ Was calculated as the total amount of the following compounds: dapagliflozin carboxylic acid (m7), hydroxy-dapagliflozin-2 (m9), and desethyl dapagliflozin coeluted in mice; dapagliflozin carboxylic acid (m7) and hydroxy-dapagliflozin-2 (m9) coeluted in rats.

⁹⁵ [REDACTED]

Table 4. Formulations used in the main clinical studies

Formulations	Development phase (Study No.)	
	Japan	Overseas
Capsules (2.5 mg, 10 mg, 100 mg)	Phase I (MB102010 ^{a)})	Phase I (MB102004 ^{a)} , D1690C00001 ^{a)})
Uncoated tablets (2.5 mg, 10 mg, 50 mg)	-	Phase I (MB102007 ^{a)} , MB102017 ^{a)} , D1690C00001 ^{a)})
Film-coated tablets (early batches) (1 mg, 2.5 mg, 5 mg, 10 mg)	Phase I (MB102025 ^{a)} , D1692C00002 ^{a)}), phase II (D1692C00005 ^{a)})	Phase I (MB102007 ^{a)} , MB102026 ^{a)} , MB102027 ^{a)} , MB102036 ^{a)} , MB102037 ^{a)} , MB102057 ^{a)} , MB102058 ^{a)} , MB102059 ^{a)} , MB102074 ^{a)} , MB102093 ^{a)}), Phase II/III (MB102029 ^{b)})
Film-coated tablets (late batches) (2.5 mg, 5 mg, 10 mg)	Phase III (D1692C00006 ^{a)} , D1692C00012 ^{a)})	-
Proposed commercial formulation (5 mg, 10 mg)	-	Phase I (MB102062 ^{b)})

a) Evaluation data, b) reference data

Human biomaterials were quantitated by LC-MS/MS or liquid chromatography/accelerator mass spectrometry (LC/AMS). The lower limit of quantitation of the LC-MS/MS method was 0.1, 1, or 10 ng/mL for unchanged dapagliflozin in plasma, 1 or 10 ng/mL for desethyl dapagliflozin in plasma, 0.2, 1, 2, or 5 ng/mL for dapagliflozin 3-O-glucuronide in plasma, 1 or 10 ng/mL for unchanged dapagliflozin in urine, and 10 ng/mL for desethyl dapagliflozin and dapagliflozin 3-O-glucuronide in urine. The lower limit of quantitation of the LC/AMS method was 0.009074 ng/mL for unchanged dapagliflozin in plasma.

Regarding biopharmaceutical studies, the results from a foreign bioavailability study (Study MB102059) were submitted as the evaluation data, and the results from 4 studies (Studies MB102005, MB102019, MB102062, and MB102090) were submitted as the reference data. Primary study results are shown below.

4.(i).A.(1) Bioavailability study (5.3.1.1.1, Study MB102059 [July to August 2009])

An open-label study was conducted to evaluate the absolute bioavailability of dapagliflozin after a single oral dose in foreign healthy adult male subjects (target sample size, 7).

A single oral dose of dapagliflozin 10 mg was to be administered under fasted conditions, and 1 hour later, ¹⁴C-dapagliflozin 80 µg was to be intravenously administered over 1 minute.

All of the 7 treated subjects were included in the safety and pharmacokinetic analysis sets.

Regarding pharmacokinetics, the geometric means (coefficient of variation [CV] %) of C_{max}, AUC_{0-t}, and AUC_{inf} of unchanged dapagliflozin after oral administration were 143 (29) ng/mL, 598 (17) ng·h/mL, and 628 (17) ng·h/mL, respectively, t_{1/2} (mean ± standard deviation [SD]) was 13.7 ± 3.44 h, and median t_{max} (minimum, maximum) was 1.03 (0.50, 1.50) h. C_{max}, AUC_{0-t}, and AUC_{inf} of unchanged dapagliflozin after intravenous administration were 10.2 (49) ng/mL, 6.43 (23) ng·h/mL, and 6.78 (22) ng·h/mL, respectively, t_{1/2} was 12.2 ± 5.25 h, and t_{max} was 0.03 (0.03, 0.08) h. The geometric mean (CV%) of CL after intravenous administration was 207 (23) mL/min, and V_{ss} (mean ± SD) was 118 ± 31.6 L. The geometric mean [two-sided 90% confidence interval (CI)] of dose-adjusted absolute bioavailability⁹⁶ of dapagliflozin was 77.8% [73.2, 82.8].

Regarding safety, 6 adverse events were reported by 3 of 7 subjects, and among these, 1 event of epistaxis in 1 subject was considered to be an adverse event for which a causal relationship to the study drug could not be ruled out (adverse drug reaction). There were no deaths, serious adverse events, or adverse events leading to treatment discontinuation. A total of 6 laboratory

⁹⁶ Absolute bioavailability = ([AUC_{inf}]_{p.o.}/dose_{p.o.})/([AUC_{inf}]_{i.v.}/dose_{i.v.})

abnormalities (all were an increase in urinary glucose) were reported by 6 of 7 subjects, but none was assessed as an adverse event.

4.(i).A.(2) Food effect study (5.3.1.2.3, Study MB102062 [April to May 2010], Reference data)

A randomized, open-label, six-treatment, three-period crossover study was conducted to evaluate the effects of a high-fat meal on the pharmacokinetics of the proposed commercial formulation and bioequivalence between the thermal-loaded⁹⁷ and non-thermal-loaded formulations of dapagliflozin in foreign healthy adult subjects (target sample size, 30).

A single oral dose of dapagliflozin 10 mg (non-thermal-loaded formulation) under fasted conditions, dapagliflozin 10 mg (thermal-loaded formulation) under fasted conditions, or dapagliflozin 10 mg (thermal-loaded formulation) of within 5 minutes after a high-fat meal was to be administered on Day 1 of each period. A washout period of ≥ 4 days was required between each period.

All of the 29 treated subjects were included in the safety and pharmacokinetic analysis sets.⁹⁸

Regarding pharmacokinetics, the ratio of geometric least-squares means (fed/fasted) [two-sided 90% CI] for AUC_{0-t} , AUC_{inf} , and C_{max} of dapagliflozin after administration of the thermal-loaded formulation was 0.961 [0.932, 0.990], 0.973 [0.943, 1.004], and 0.550 [0.499, 0.606], respectively. The ratio of geometric least-squares means (thermal-loaded formulation/non-thermal-loaded formulation) [two-sided 90% CI] for AUC_{0-t} , AUC_{inf} , and C_{max} of dapagliflozin after administration under fasted conditions was 0.988 [0.959, 1.018], 0.990 [0.960, 1.021], and 1.018 [0.923, 1.122], respectively.

Regarding safety, 14 adverse events were reported by 7 of 29 subjects. Of these, 13 events in 7 subjects were classified as adverse drug reaction; 1 event in 1 subject who received the non-thermal-loaded formulation under fasted conditions (diarrhoea), 1 event in 1 subject who received the thermal-loaded formulation under fasted conditions (diarrhoea), and 11 events in 6 subjects who received the thermal-loaded formulation after a meal (headache in 2 subjects, fatigue/pain/nausea/retching/feeling of body temperature change, nausea/flatulence, hypersensitivity, and fatigue in 1 subject each) were included. There were no deaths, serious adverse events, or adverse events leading to treatment discontinuation.

4.(ii) Summary of clinical pharmacology studies

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

As the evaluation data, the results from the following studies were submitted: 3 Japanese phase I studies (MB102010, MB102025, and D1692C00002); and 13 foreign phase I studies (MB102004, MB102006, MB102007, MB102017, MB102026, MB102027, MB102036, MB102037, MB102057, MB102058, MB102074, MB102093, and D1690C00001). As the reference data, the results from 5 foreign phase I studies (MB102001, MB102002, MB102003, MB102066, and MB102088) were submitted. Primary study results are shown below.

4.(ii).A.(1) Studies using human biomaterials (4.2.2.2.1, 4.2.2.4.1, 4.2.2.4.2, 4.2.2.3.1, 4.2.2.7.1, 5.3.2.1.3, 5.3.2.2.1 to 5.3.2.2.8, 5.3.2.3.1, 5.3.2.3.3)

The mean plasma protein binding (equilibrium dialysis method) of dapagliflozin (0.5, 5 μ g/mL) and dapagliflozin 3-O-glucuronide (the major human metabolite) (0.5, 5 μ g/mL) was 91% and

⁹⁷ [REDACTED]

⁹⁸ One subject who discontinued the study on his/her own decision before the start of Period 3 (treatment period with the non-thermal-loaded formulation) was excluded from the pharmacokinetic analysis of the non-thermal-loaded formulation.

89%, respectively. The mean percent distribution in red blood cells was 37% in human fresh blood containing dapagliflozin at 5 μ M.

The membrane permeability of dapagliflozin (2.7-50.5 μ M) was determined using Caco-2 cells at pH 7.4. The results showed that the permeability coefficient (P_c) in the absorptive direction (from the apicolateral surface to the basolateral surface [A to B]) was 37 to 43 nm/sec and the P_c in the secretory direction (from the basolateral surface to the apicolateral surface [B to A]) was 120 to 138 nm/sec. The ratio ($P_{c\ A\ to\ B}/P_{c\ B\ to\ A}$) was found to be in the range of 2.9 to 3.8 and decreased to 1.5 to 1.7 in the presence of ketoconazole or cyclosporin A, i.e., inhibitors of P-glycoprotein (P-gp); therefore, dapagliflozin was considered to be a substrate of P-gp. The membrane permeability by passive diffusion was found to be high in the parallel artificial membrane permeability assay (P_c [nm/sec], 211 [pH 5.5], 238 [pH 7.4]). In addition, the membrane permeability of digoxin (a substrate of P-gp) was evaluated in the presence and absence of dapagliflozin (0.19-57.6 μ M) and dapagliflozin 3-O-glucuronide (0.14-20.1 μ M) using Caco-2 cells. The results showed no inhibitory activities against P-gp (IC_{50} , >57.6 and >20.1 μ M, respectively).

After incubation of human liver microsomes with dapagliflozin (3, 10 μ M) in the presence of NADPH or UDPGA, the metabolic rates were 120 and 60 pmol/min/mg protein, respectively. After incubation of human liver, kidney, and intestinal microsomes with dapagliflozin (100 μ M) in the presence of UDPGA, the formation rates of dapagliflozin 2-O-glucuronide were 2.80, 1.34, and 0.81 pmol/min/mg protein, respectively, and those of dapagliflozin 3-O-glucuronide were 60.8, 184, and 1.69 pmol/min/mg protein, respectively. The intrinsic clearance (V_{max}/K_m) of dapagliflozin 3-O-glucuronidation (1.66 μ L/min/mg protein) was faster than that of dapagliflozin 2-O-glucuronidation (0.0173 μ L/min/mg protein) in human kidney microsomes. From a study using human liver microsomes stratified by genotype, rare allelic variations UGT1A9*1*3 and UGT1A9*3*3 were suggested to be involved in the interpatient variability in exposure.

CYP isoforms⁹⁹ involved in the metabolism of dapagliflozin (1-100 μ M) were investigated using CYP expression systems in the presence of NADPH. As a result, CYP1A2, CYP2C9, CYP2D6, and CYP3A4 showed relatively higher metabolic activities¹⁰⁰ than other CYP isoforms. The uridine diphosphate-glucuronosyltransferase (UGT) isoforms¹⁰¹ involved in the glucuronidation of dapagliflozin (5, 100 μ M) were investigated using human UGT isoforms in the presence of UDPGA. The results showed an involvement of UGT1A9, as evidenced by the highest concentrations of dapagliflozin 3-O-glucuronide with UGT1A9. Human liver microsomes were inhibited from forming dapagliflozin 3-O-glucuronide by mefenamic acid¹⁰² (IC_{50} , 1.17 μ M) and niflumic acid¹⁰³ (IC_{50} , 0.091 μ M); similar results were obtained in a study using recombinant UGT1A9. In addition, niflumic acid inhibited human kidney microsomes from forming dapagliflozin 3-O-glucuronide (IC_{50} , 0.4 μ M).

Using human liver microsomes and human hepatocytes, metabolites of ^{14}C -dapagliflozin (10 μ M in liver microsomes, 30 μ M in hepatocytes) were investigated. The results showed that the main metabolites were benzylic hydroxy-dapagliflozin and desethyl dapagliflozin in human liver microsomes and dapagliflozin 3-O-glucuronide in human hepatocytes, but no human-specific metabolites were detected.

⁹⁹ CYP1A1, 1A2, 1B1, 2A6, 2B6, 2C8, 2C9, 2C19, 2D6, 2E1, 3A4, 3A5, and 4A11 were examined.

¹⁰⁰ Dapagliflozin (100 μ M) was metabolized by CYP2D6 at a rate of 5.74 pmol/min/pmol CYP, by CYP1A2 at a rate of 4.40 pmol/min/pmol CYP, by CYP3A4 at a rate of 4.05 pmol/min/pmol CYP, by CYP2C9 at a rate of 3.87 pmol/min/pmol CYP, and by other isoforms at a rate of \leq 2.2 pmol/min/pmol CYP.

¹⁰¹ UGT1A1, 1A3, 1A4, 1A6, 1A7, 1A8, 1A9, 1A10, 2B4, 2B7, 2B15, and 2B17 were examined.

¹⁰² An inhibitor of UGT1A9 and UGT2B7

¹⁰³ A specific inhibitor of UGT1A9

In Study MB102006, dapagliflozin 3-*O*-glucuronide (approximately 80% of the total urinary radioactivity) and dapagliflozin 2-*O*-glucuronide (approximately 5% of the total urinary radioactivity) were detected in human urine collected for 12 hours post-dose.

Inhibition of CYP isoforms (CYP1A2, 2C9, 2C19, 2D6, and 3A4) by dapagliflozin or desethyl dapagliflozin was evaluated using human CYP-expressing microsomes in the presence of NADPH. The results showed that IC₅₀ of dapagliflozin was >40 µM and that of desethyl dapagliflozin was >100 µM for all the isoforms. Similarly, inhibition of CYP isoforms (CYP1A2, 2A6, 2B6, 2C8, 2C9, 2C19, 2D6, and 3A4) by dapagliflozin was evaluated using human liver microsomes, and the results showed that IC₅₀ of dapagliflozin was >45 µM for all the isoforms, with no time-dependent inhibition detected.

Induction of CYP isoforms (CYP1A2, 2B6, and 3A4/5) by dapagliflozin was evaluated using human hepatocytes, and the results showed no induction up to 20 µM of dapagliflozin.

Human liver microsomes were incubated with dapagliflozin (0.0122-50 µM) and β-estradiol (30 µM) in the presence of UDPGA and inhibition against UGT1A1 was evaluated. The results showed no inhibition against UGT1A1 (IC₅₀ >50 µM).

Uptake of dapagliflozin or dapagliflozin 3-*O*-glucuronide (both at 0.25-100 µM) was evaluated using human organic anion transporter (hOAT) 1-expressing MDCK cells or hOAT3- or human organic cation transporter (hOCT) 2-expressing HEK-293 cells. The results suggested that dapagliflozin and dapagliflozin 3-*O*-glucuronide are substrates of hOAT3 (IC₅₀; 33 and 100 µM, respectively). In addition, uptake of dapagliflozin (0.02-100 µM) was evaluated using human organic anion-transporting polypeptide (hOATP) 1B1- or hOATP1B3-expressing HEK-293 cells, and the results showed that the IC₅₀ value was 69.3 or 8.0 µM, respectively.

Transcriptional activation of pregnane X receptor (PXR) by dapagliflozin (0.1-25 µM) was evaluated using human PXR-expressing HepG2/C3A cells. The results showed that dapagliflozin did not activate human PXR at up to 25 µM.

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

4.(ii).A.(2).1 Ascending, single-dose study in Japanese healthy adult subjects (5.3.3.1.1, Study MB102010 [June to August 2007])

A randomized, double-blind, placebo-controlled study with a dose escalation design was conducted to evaluate the safety, tolerability, pharmacokinetics, and pharmacodynamic effects of a single oral dose of dapagliflozin in Japanese healthy adult male subjects (target sample size, 32).

A single oral dose of placebo or dapagliflozin at a dose of 2.5, 10, 20, or 50 mg was to be administered under fasted conditions in Steps 1 to 4. For each Step, 2 subjects and 6 subjects, respectively, were randomly assigned to placebo and dapagliflozin groups.

All of the 32 treated subjects were included in the safety and pharmacodynamic analysis sets, and 24 subjects who received dapagliflozin were included in the pharmacokinetic analysis set.

Regarding pharmacokinetics, the pharmacokinetic parameters of unchanged dapagliflozin and dapagliflozin 3-*O*-glucuronide after single oral administration of dapagliflozin were as shown in Table 5. The geometric mean (CV%) of the molar ratio¹⁰⁴ of exposure (AUC_{inf}) (dapagliflozin 3-*O*-glucuronide/unchanged dapagliflozin) was 1.30 (46), 1.15 (38), 1.14 (33), and 1.52 (18), respectively, in the dapagliflozin 2.5, 10, 20, and 50 mg groups.

¹⁰⁴ Molar ratio = (AUC_{inf} of dapagliflozin 3-*O*-glucuronide/AUC_{inf} of unchanged dapagliflozin) × (molecular weight of unchanged dapagliflozin/molecular weight of dapagliflozin 3-*O*-glucuronide)

Table 5. Pharmacokinetic parameters of unchanged dapagliflozin and dapagliflozin 3-O-glucuronide after single oral administration of dapagliflozin

Analyte	Dose (mg)	No. of subjects	C _{max} (ng/mL)	t _{max} (h)	AUC _{0-t} (ng·h/mL)	AUC _{inf} (ng·h/mL)	t _{1/2} (h)	CL _R (mL/min)	Urinary excretion rate ^{a)} (%)
Unchanged dapagliflozin	2.5	6	29 (14)	1.00 (1.00, 2.00)	89 (31)	103 (30)	8.1 ± 4.78	3.9 ± 0.86	1.0 ± 0.48
	10	6	124 (34)	1.25 (1.00, 1.50)	464 (20)	489 (19)	12.1 ± 7.79	3.6 ± 0.91	1.1 ± 0.38
	20	6	265 (26)	1.00 (0.50, 2.00)	915 (15)	939 (14)	12.2 ± 4.70	2.7 ± 0.52	0.8 ± 0.14
	50	6	610 (22)	1.25 (1.00, 1.50)	2058 (24)	2093 (24)	12.1 ± 7.03	3.6 ± 1.23	0.9 ± 0.24
Dapagliflozin 3-O-glucuronide	2.5	6	46 (49)	1.75 (1.00, 2.00)	170 (31)	191 (27)	11.9 ± 6.19	125 ± 36.2	38.8 ± 2.31
	10	6	199 (34)	2.00 (1.00, 2.00)	778 (32)	804 (32)	11.8 ± 4.42	106 ± 30.3	34.6 ± 4.32
	20	6	367 (45)	1.50 (1.00, 4.00)	1506 (34)	1535 (36)	10.0 ± 3.29	93 ± 26.5	28.9 ± 2.23
	50	6	1144 (15)	1.50 (1.00, 2.00)	4495 (23)	4550 (22)	12.0 ± 4.67	94 ± 19.2	35.6 ± 5.43

C_{max} and AUC are expressed in the geometric mean (CV%), t_{max} in median (minimum, maximum), and the other data in mean ± SD.

C_{max}, Maximum plasma concentration; t_{max}, Time to reach the maximum plasma concentration; AUC_{0-t}, Area under the plasma concentration-time curve from 0 to the last sampling point (t); AUC_{inf}, Area under the plasma concentration-time curve from 0 to infinity; t_{1/2}, Elimination half-life; CL_R, Renal clearance

a) Urinary excretion rate of dapagliflozin 3-O-glucuronide = (urinary excretion of dapagliflozin 3-O-glucuronide/dose of dapagliflozin) × (molecular weight of unchanged dapagliflozin/molecular weight of dapagliflozin 3-O-glucuronide)

Regarding pharmacodynamics, the 120-hour cumulative urinary glucose excretion (mean ± SD) after administration of the study drug was 0.34 ± 0.04, 37.56 ± 14.76, 97.52 ± 29.27, 110.17 ± 16.63, and 143.70 ± 21.64 g, respectively, in the placebo, dapagliflozin 2.5, 10, 20, and 50 mg groups. The blood glucose AUC_{0-4h} (mean ± SD) for up to 4 hours after the start of midday meal was 436.94 ± 56.20, 483.04 ± 19.22, 465.17 ± 32.71, 473.21 ± 28.46, and 466.65 ± 41.10 mg·h/dL, respectively.

Regarding safety, 27 adverse events were reported by 24 of 32 subjects. Six events (all are glucose urine present) reported by 6 of 6 subjects each in the dapagliflozin 2.5, 10, 20, and 50 mg groups were classified as adverse drug reactions, but all were mild in severity and resolved within 1 to 3 days after onset. There were no deaths, serious adverse events, or adverse events leading to treatment discontinuation.

4.(ii).A.(2).2 Mass balance study (5.3.3.1.4, Study MB102006 [September to October 2005])

An open-label, uncontrolled study was conducted to evaluate the pharmacokinetics and safety of a single oral dose of ¹⁴C-dapagliflozin in foreign healthy adult male subjects (target sample size, 6).

A single oral dose of ¹⁴C-dapagliflozin 50 mg (solution formulation) was to be administered under fasted conditions.

All of the 6 treated subjects were included in the safety and pharmacokinetic analysis sets.

Regarding pharmacokinetics, the pharmacokinetic parameters of unchanged dapagliflozin, desethyl dapagliflozin, and the total radioactivity (in plasma and in whole blood) after administration of ¹⁴C-dapagliflozin were as shown in Table 6. The geometric mean (CV%) of the

molar ratio¹⁰⁵ of exposure (AUC_{inf}) (desethyl dapagliflozin/unchanged dapagliflozin) was 0.01 (12.55). The geometric mean ratio (whole blood/plasma) for AUC_{inf} of total radioactivity was 0.58.

Table 6. Pharmacokinetic parameters of unchanged dapagliflozin, desethyl dapagliflozin, and the total radioactivity (in plasma and in whole blood) after single oral administration of ^{14}C -dapagliflozin

Analyte	No. of subjects	C_{max} (ng/mL)	t_{max} (h)	AUC_{inf} (ng·h/mL)	$t_{1/2}$ (h)	CL_R (mL/min)	Urinary excretion rate (%)	Fecal excretion rate (%)
Unchanged dapagliflozin	6	550 (18)	0.50 (0.50, 0.75)	2426 (26)	13.77 ± 9.36	6.88 ± 4.67	1.61 ± 0.83	-
Desethyl dapagliflozin	6	6 (24)	0.75 (0.75, 0.75)	16 (28)	2.47 ± 1.22	45.54 ± 24.89	0.07 ± 0.04	-
Total radioactivity ^{a)} (plasma)	6	1761 (16)	1.00 (0.75, 1.00)	6952 (22)	5.39 ± 2.86	107.73 ± 22.01	75.16 ± 9.19	20.99 ± 8.81
Total radioactivity ^{a)} (whole blood)	6	1105 (17)	0.88 (0.75, 1.00)	4029 (19)	3.95 ± 1.66	-	-	-

C_{max} and AUC are expressed in the geometric mean (CV%), t_{max} in median (minimum, maximum), and the other data in mean ± SD.

C_{max} , Maximum plasma concentration; t_{max} , Time to reach the maximum plasma concentration; AUC_{inf} , Area under the plasma

concentration-time curve from 0 to infinity; $t_{1/2}$, Elimination half-life; CL_R , Renal clearance; -, Not applicable

a) The unit of total radioactivity in plasma and whole blood is ng eq/mL for C_{max} and ng eq·h/mL for AUC_{inf} .

Regarding safety, no adverse events were reported. Regarding laboratory parameters, increased serum creatinine (1.33-fold increase) was reported by 1 of 6 subjects, but this variability fell within the normal limits.

4.(ii).A.(3) Studies in patients

4.(ii).A.(3).1 Ascending multiple-dose study in Japanese patients with type 2 diabetes mellitus (5.3.3.2.1, Study MB102025 [November 2007 to May 2008])

A randomized, double-blind, placebo-controlled study with a dose escalation design was conducted to evaluate the safety, tolerability, pharmacokinetics, and pharmacodynamic effects of multiple oral doses of dapagliflozin in Japanese patients with type 2 diabetes mellitus (target sample size, 36).

Placebo or dapagliflozin at a dose of 2.5, 10, or 20 mg was to be administered orally once daily in the morning for 14 days in Steps 1 to 3. For each Step, 3 subjects and 9 subjects were randomly assigned to placebo and dapagliflozin groups, respectively.

All of the 36 treated subjects were included in the safety and pharmacodynamic analysis sets, and 27 subjects who received dapagliflozin were included in the pharmacokinetic analysis set.

Regarding pharmacokinetics, the pharmacokinetic parameters of unchanged dapagliflozin and dapagliflozin 3-O-glucuronide after multiple oral administration of dapagliflozin were as shown in Table 7. The geometric mean (CV%) of the molar ratio¹⁰⁶ of exposure (AUC_{τ}) (dapagliflozin 3-O-glucuronide/unchanged dapagliflozin) on Day 1 was 2.20 (21), 1.74 (33), and 2.20 (18), respectively, in the dapagliflozin 2.5, 10, and 20 mg groups, and that on Day 14 was 1.93 (17), 1.52 (33), and 1.98 (17), respectively.

¹⁰⁵ Molar ratio = (AUC_{inf} of desethyl dapagliflozin/ AUC_{inf} of unchanged dapagliflozin) × (molecular weight of unchanged dapagliflozin/molecular weight of desethyl dapagliflozin)

¹⁰⁶ Molar ratio = (AUC_{τ} of dapagliflozin 3-O-glucuronide/ AUC_{τ} of unchanged dapagliflozin) × (molecular weight of dapagliflozin/molecular weight of dapagliflozin 3-O-glucuronide)

Table 7. Pharmacokinetic parameters of unchanged dapagliflozin and dapagliflozin 3-O-glucuronide after multiple oral administration of dapagliflozin^{a)}

Analyte	Dose (mg)	No. of subjects	Day	C _{max} (ng/mL)	t _{max} (h)	AUC _τ (ng·h/mL)	CL _R (mL/min)	Accumulation ratio	Urinary excretion rate (%)
Unchanged dapagliflozin	2.5	9	Day 1	43 (30)	0.50 (0.50, 1.00)	123 (29)	3.82 ± 1.064	-	1.17 ± 0.417
			Day 14	48 (27)	0.50 (0.50, 1.00)	157 (27)	4.59 ± 1.946	1.28 (11)	1.68 ± 0.549
	10	9	Day 1	188 (27)	1.00 (0.50, 1.00)	602 (23)	3.75 ± 0.911	-	1.36 ± 0.331
			Day 14	191 (35)	1.00 (0.50, 1.50)	727 (23)	4.30 ± 1.005	1.21 (7)	1.90 ± 0.587
	20	9	Day 1	298 (21)	1.00 (0.50, 2.00)	1027 (15)	4.44 ± 0.577	-	1.38 ± 0.259
			Day 14	305 (31)	1.00 (0.50, 2.00)	1225 (17)	5.10 ± 1.445	1.19 (4)	1.89 ± 0.572
Dapagliflozin 3-O-glucuronide	2.5	9	Day 1	99 (22)	1.00 (1.00, 1.50)	388 (21)	135 ± 45.8	-	85 ± 6.9
			Day 14	97 (21)	1.00 (1.00, 1.50)	435 (21)	146 ± 51.8	1.12 (11)	102 ± 10.2
	10	9	Day 1	359 (23)	1.00 (1.00, 1.50)	1494 (26)	138 ± 28.8	-	85 ± 12.6
			Day 14	316 (25)	1.02 (1.00, 2.00)	1581 (32)	152 ± 33.0	1.06 (14)	100 ± 16.1
	20	9	Day 1	765 (25)	1.00 (1.00, 4.00)	3239 (17)	130 ± 17.2	-	88 ± 9.8
			Day 14	698 (20)	1.50 (1.00, 4.00)	3466 (17)	133 ± 27.6	1.07 (6)	96 ± 15.5

C_{max}, AUC, and accumulation ratio are expressed in the geometric mean (CV%), t_{max} in median (minimum, maximum), and the other data in mean ± SD.

C_{max}, Maximum plasma concentration; t_{max}, Time to reach the maximum plasma concentration; AUC_τ, Area under the concentration-time curve at dosing interval at steady state; t_{1/2}, Elimination half-life; CL_R, Renal clearance; -, Not applicable

a) t_{1/2} was exploratorily calculated because sampling intervals were inadequate for determination of terminal phase (t_{1/2} on Day 14 after multiple administration of dapagliflozin at doses of 2.5-20 mg were approximately █-█ hours for unchanged dapagliflozin and █-█ hours for dapagliflozin 3-O-glucuronide).

Regarding pharmacodynamics, the 24-hour cumulative urinary glucose excretion (mean ± SD) after administration of the study drug on Day 1 was 9.369 ± 15.429, 37.852 ± 12.806, 68.374 ± 13.439, and 76.681 ± 18.073 g in the placebo, dapagliflozin 2.5, 10, and 20 mg groups, respectively, and that on Day 14 was 6.833 ± 11.396, 41.626 ± 13.399, 71.443 ± 11.423, and 73.026 ± 20.822 g, respectively. The absolute change from baseline in the 24-hour cumulative urinary Ca excretion (creatinine-corrected value) after administration of dapagliflozin was 8 to 22 mg (0.00-0.02 mg Calcium/mg Cr) on Day 1 and 10 to 21 mg (0.01 mg Calcium/mg Cr) on Day 14, with no dose-dependent changes observed. The geometric mean ratios (Day 13 to 2 days before administration) for AUC_{0-4h} of serum glucose and insulin levels after oral glucose tolerance test (OGTT), conducted 2 days before administration and on Day 13, were 0.849 to 0.912 and 0.726 to 0.840, respectively, in the dapagliflozin group. On Days 1 and 14, the serum glucose, insulin, and C-peptide levels all decreased; the maximum percentage changes from baseline in serum glucose across all dapagliflozin groups were in the ranges of -15.6% to -10.4% and -21.9% to -15.2%, respectively, those in serum insulin were in the ranges of -34.4% to -27.2% and -40.0% to -35.9%, respectively, and those in C-peptide were in the ranges of -23.9% to -17.5% and -50.9% to -30.0%, respectively. The inhibition rates of glucose reabsorption before and after administration of the study drug were as shown in Table 8.

Table 8. Inhibition rates of glucose reabsorption before and after administration of the study drug

	Dose	1 day before treatment	Day 1	Day 14
Between 0-4 hours post-dose (%)	Placebo	0.549 ± 1.278	0.296 ± 0.648	0.520 ± 1.241
	2.5 mg	0.169 ± 0.376	21.211 ± 3.992	26.434 ± 7.374
	10 mg	0.131 ± 0.159	33.776 ± 3.891	41.994 ± 7.616
	20 mg	0.100 ± 0.052	35.361 ± 7.113	42.224 ± 11.571
Between 4-8 hours post-dose (%)	Placebo	0.175 ± 0.339	0.183 ± 0.360	0.197 ± 0.368
	2.5 mg	0.066 ± 0.091	17.752 ± 5.517	17.584 ± 5.955
	10 mg	0.056 ± 0.022	35.171 ± 8.285	34.859 ± 9.452
	20 mg	0.060 ± 0.027	34.484 ± 6.264	32.756 ± 4.717

Mean ± SD, n = 9

Regarding safety, 22 adverse events were reported by 16 of 36 subjects. One adverse drug reaction (protein urine present) occurred in 1 of 9 subjects in the placebo group, 4 adverse drug reactions in 4 of 9 subjects in the 2.5 mg group (2 events of protein urine present, 1 event each of thirst and pollakiuria), 2 adverse drug reactions in 2 of 9 subjects in the 10 mg group (blood alkaline phosphatase [ALP] increased, nocturia), and 2 adverse drug reactions in 2 of 9 subjects in the 20 mg group (protein urine present). Protein urine present in 1 subject in the 20 mg group was not found to be reversible. There were no deaths, serious adverse events, or adverse events leading to treatment discontinuation. Abnormal values (2 events of blood ALP increased, 1 event each of blood lactate dehydrogenase increased, blood potassium [K] increased, blood urea increased, and bacteriuria) were found in laboratory testing (in blood and urine¹⁰⁷), but these values returned to normal without treatment and therefore, were not considered clinically significant. No changes of specific concern were seen in vital signs or electrocardiogram (ECG).

4.(ii).A.(3).2 Population pharmacokinetic analysis (5.3.3.5.1, Studies MB102025 and D1692C00005)

In order to identify covariates that affect the pharmacokinetics of dapagliflozin using 2198 measured values of plasma concentration of unchanged dapagliflozin obtained from Japanese clinical studies (Studies MB102025 and D1692C00005) in patients with type 2 diabetes mellitus who have normal renal function or mild renal impairment, a population pharmacokinetic analysis was performed using a nonlinear mixed-effects modeling (NONMEM software [version 7.2.0]) based on a 2-compartment model. The analysis data set included 251 subjects (195 males, 56 females) with age (median [minimum-maximum]) of 59 [30-77], body weight of 66.3 [39.5-126] kg, and eGFR of 81.67 [48.61-135.64] mL/min/1.73 m². Based on the results of analysis using forward selection and backward elimination methods of covariates¹⁰⁸ for oral clearance (CL/F), eGFR, body weight, and sex were included in the final model as covariates. From the final model, a change in eGFR from 81.67 to 113.18 mL/min/1.73 m² was estimated to lead to an increase in CL/F by 9%, and a change in eGFR from 81.67 to 63.31 mL/min/1.73 m² to a decrease in CL/F by 6.7%. A change in body weight from 66.3 to 91.8 kg was estimated to lead to an increase in CL/F by 15%, and a change in body weight from 66.3 to 49.2 kg to a decrease in CL/F by 11.9%. CL/F in female subjects was estimated to be lower by 14.2% than that in male subjects.

¹⁰⁷ Measurements of uric acid as a urinary safety variable showed a tendency for uric acid excretion to be increased in the dapagliflozin groups compared to the placebo group (the change from baseline [mean ± SD] in 24-hour cumulative urinary uric acid excretion after administration of the study drug on Day 1 was 48 ± 70, 76 ± 33, 108 ± 66, and 124 ± 69 mg in the placebo, dapagliflozin 2.5 mg, 10 mg, and 20 mg groups, respectively, and that on Day 14 was 20 ± 50, 4 ± 46, 19 ± 63, and 47 ± 83 mg, respectively).

¹⁰⁸ The examined covariates include study, age, sex, body weight, ideal body weight, BMI, creatinine clearance, eGFR, albumin, total bilirubin, aspartate aminotransferase, alanine aminotransferase, and ALP.

4.(ii).A.(4) Studies on intrinsic factor

4.(ii).A.(4).1 Study in patients with renal impairment (5.3.3.3.1, Study MB102007 [March 2006 to October 2008])

An open-label, parallel-group study was conducted to evaluate the pharmacodynamic effects, pharmacokinetics, and safety of dapagliflozin in foreign healthy adult subjects and patients with type 2 diabetes mellitus who have normal renal function or renal impairment (target sample size; 38 subjects in Period I, 18 subjects in Period II).

In Period I, a single oral dose of dapagliflozin 50 mg was to be administered to healthy adult subjects (8 subjects; Creatinine clearance (CLcr), >80 mL/min), patients with normal renal function (12 subjects; CLcr, >80 mL/min), patients with mild renal impairment (8 subjects; >50 and ≤80 mL/min), patients with moderate renal impairment (8 subjects; >30 and ≤50 mL/min), and patients with severe renal impairment (4 subjects; CLcr, <30 mL/min). In Period II, that followed a 48-hour washout period, oral administration of dapagliflozin at a dose of 20 mg was to be given once daily for 7 days in the groups of patients with type 2 diabetes mellitus (4 subjects each in the normal renal function and mild renal impairment groups, 7 subjects in the moderate renal impairment group, 3 subjects in the severe renal impairment group).

All of the 40 treated subjects were included in the safety, pharmacokinetic,¹⁰⁹ and pharmacodynamic¹¹⁰ analysis sets. Two subjects discontinued the study during Period I (the moderate renal impairment group, 1 patient for consent withdrawal; the severe renal impairment group, 1 patients for adverse event), and 2 subjects discontinued the study during Period II (the moderate renal impairment group, 1 patient each for consent withdrawal and adverse event).

Regarding pharmacokinetics, the pharmacokinetic parameters of unchanged dapagliflozin and dapagliflozin 3-O-glucuronide after single and multiple oral administration¹¹¹ of dapagliflozin were as shown in Table 9 and Table 10, respectively.

¹⁰⁹ One patient in the severe renal impairment group who discontinued the study during Period I due to an adverse event was excluded from the analysis sets for AUC_{inf} and t_{1/2}. One patient in the mild renal impairment group who had an abnormally high blood concentration at pre-dose on Day 5 of Period II was included in the pharmacokinetic analysis set using measurements of unchanged dapagliflozin and the metabolites determined at 2 separate institutions with correction of the analytical errors.

¹¹⁰ Timepoints when blood was collected under non-fasted conditions (4 subjects, 2 patients each in the normal renal function group and the moderate renal impairment group) and a timepoint with missing data (1 patient in the severe renal impairment group) were excluded from the analysis sets for fasting blood glucose. In addition, 4 subjects from whom 24-hour urine collection was inappropriately performed (2 patients each in the normal renal function group and the moderate renal impairment group) were excluded from the analysis sets for 24-hour cumulative urinary glucose excretion after administration of dapagliflozin on the relevant days. Seven subjects for whom urinary glucose excretion up to 6 hours post-dose or blood glucose AUC_{0-6h} could not be calculated were excluded from the analysis set for glucose clearance on the relevant days.

¹¹¹ Regression analysis of parameters after multiple administration was performed using log-transformed C_{max} and AUC_T on Days 4 and 10.

Table 9. Pharmacokinetic parameters of unchanged dapagliflozin and dapagliflozin 3-O-glucuronide after single oral administration of dapagliflozin

Analyte	Study population	Severity of renal impairment	No. of subjects	C _{max} (ng/mL)	t _{max} (h)	AUC _{0-t} (ng·h/mL)	AUC _{inf} (ng·h/mL)	t _{1/2} (h)	CL _R (mL/min)	Urinary excretion rate (%)
Unchanged dapagliflozin	Healthy adult subjects Patients with type 2 diabetes mellitus	Normal	8	710 (31)	1.17 (0.50, 2.00)	2821 (27)	2880 (27)	12.7 ± 6.95	3.52 (34)	1.2 ± 0.45
		Normal	12	647 (37)	1.25 (0.50, 2.00)	2439 (30)	2504 (30)	11.9 ± 5.72	3.43 (47)	1.0 ± 0.44
		Mild	8	902 (35)	1.25 (0.50, 2.00)	3832 (26)	4018 (26)	18.4 ± 8.15	2.85 (55)	1.4 ± 0.96
		Moderate	8	897 (41)	1.00 (0.50, 2.98)	4847 (35)	5182 (38)	17.9 ± 3.39	2.06 (81)	1.6 ± 1.30
		Severe	4	772 (11)	1.17 (0.75, 1.50)	4385 (12)	4884 (10) ^a	15.0 ± 4.15 ^a	0.84 (46)	0.5 ± 0.23
Dapagliflozin 3-O-glucuronide	Healthy adult subjects Patients with type 2 diabetes mellitus	Normal	8	960 (33)	2.00 (1.00, 3.00)	4565 (30)	4634 (30)	12.1 ± 5.99	151 (54)	63.1 ± 24.72
		Normal	12	1391 (29)	2.00 (1.00, 3.00)	6500 (33)	6616 (32)	11.2 ± 3.71	76 (67)	51.5 ± 30.50
		Mild	8	1641 (26)	2.00 (1.00, 3.00)	9413 (23)	9830 (24)	17.5 ± 8.17	75 (33)	64.3 ± 24.75
		Moderate	8	2037 (31)	1.75 (1.00, 3.00)	15,943 (35)	16,831 (36)	16.0 ± 2.68	43 (62)	62.6 ± 25.22
		Severe	4	1986 (22)	2.00 (1.83, 4.00)	19,045 (36)	22,409 (31) ^a	12.9 ± 2.43 ^a	11 (23)	18.5 ± 7.78

C_{max}, AUC_{0-t}, AUC_{inf}, and CL_R are expressed in the geometric mean (CV%), t_{max} in median (minimum, maximum), and t_{1/2} and urinary excretion rate in mean ± SD.

C_{max}, Maximum plasma concentration; t_{max}, Time to reach the maximum plasma concentration; AUC_{0-t}, Area under the plasma concentration-time curve from 0 to the last sampling point (t); AUC_{inf}, Area under the plasma concentration-time curve from 0 to infinity; t_{1/2}, Elimination half-life; CL_R, Renal clearance

a) n = 3

Table 10. Pharmacokinetic parameters of unchanged dapagliflozin and dapagliflozin 3-O-glucuronide after multiple oral administration of dapagliflozin in patients with type 2 diabetes mellitus

Analyte	Severity of renal impairment	No. of subjects	Day	C _{max} (ng/mL)	t _{max} (h)	AUC _τ (ng·h/mL)	CL _R (mL/min)	Urinary excretion rate (%)
Unchanged dapagliflozin	Normal	4	Day 4	249 (21)	1.00 (0.50, 1.50)	864 (11)	5.22 (38)	1.4 ± 0.51
		4	Day 10	310 (22)	1.00 (0.50, 1.00)	853 (8)	6.51 (34)	1.7 ± 0.52
	Mild	4	Day 4	410 (23)	1.00 (1.00, 6.00)	1428 (38)	2.37 (54)	1.1 ± 0.38
		4	Day 10	358 (25)	1.00 (1.00, 1.50)	1443 (21)	2.52 (51)	1.2 ± 0.54
	Moderate	6	Day 4	466 (21)	1.00 (0.50, 1.00)	1807 (31)	2.54 (71)	2.0 ± 2.15
		5	Day 10	512 (23)	1.00 (0.50, 1.50)	2467 (37)	2.06 (77)	1.8 ± 1.11
Dapagliflozin 3-O-glucuronide	Severe	3	Day 4	330 (6)	1.00 (0.50, 2.00)	1920 (26)	1.42 (30)	0.9 ± 0.43
		3	Day 10	338 (16)	1.00 (0.50, 1.00)	2207 (27)	1.13 (19)	0.8 ± 0.31
	Normal	4	Day 4	644 (21)	1.75 (1.00, 2.00)	2818 (40)	139 (48)	79.9 ± 15.41
		4	Day 10	637 (26)	1.50 (1.00, 1.50)	2769 (36)	93 (66)	60.5 ± 33.26
	Mild	4	Day 4	791 (31)	1.75 (1.50, 6.00)	4164 (22)	79 (31)	68.7 ± 20.94
		4	Day 10	843 (18)	1.50 (1.50, 2.00)	4278 (25)	72 (49)	68.0 ± 27.60
	Moderate	6	Day 4	911 (38)	1.50 (1.00, 2.00)	6502 (39)	37 (52)	50.1 ± 15.68
		5	Day 10	1088 (24)	1.50 (1.00, 2.00)	8442 (40)	37 (48)	66.0 ± 20.57
	Severe	3	Day 4	770 (21)	2.00 (1.50, 4.00)	7947 (46)	15 (39)	27.7 ± 15.97
		3	Day 10	965 (18)	2.00 (1.50, 4.00)	10,549 (31)	10 (28)	23.2 ± 9.79

C_{max}, AUC_τ, and CL_R are expressed in the geometric mean (CV%), t_{max} in median (minimum, maximum), and t_{1/2} in mean ± SD.

C_{max}, Maximum plasma concentration; t_{max}, Time to reach the maximum plasma concentration; AUC_τ, Area under the concentration-time curve at dosing interval at steady state; CL_R, Renal clearance

The results of regression analysis between iohexol clearance¹¹² and log-transformed C_{max}, AUC_{inf}, AUC_{0-t}, and AUC_τ values of unchanged dapagliflozin and dapagliflozin 3-O-glucuronide

¹¹² Between Day -16 and Day -9, 5 mL of Iohexol injection (containing 300 mg/mL of iodine) was administered as a single intravenous infusion over 15 minutes, and plasma iohexol clearance (= dose level of iohexol/plasma AUC_{inf} of iohexol) was calculated. The

after single oral administration and multiple oral administration¹¹³ of dapagliflozin showed that the two-sided 95% CI of the slope of the regression line contained zero for C_{max} on Days 4 and 10 of multiple oral administration, but was negative and did not contain zero for the other AUCs and C_{maxs} . Based on this regression analysis, a comparison of pharmacokinetic parameters after single and multiple oral administration of dapagliflozin between subjects with normal renal function (healthy adult subjects, patients with type 2 diabetes mellitus) and patients with type 2 diabetes mellitus who have renal impairment was performed. The results were as shown in Table 11.

Table 11. Comparison of pharmacokinetic parameters after single and multiple oral administration of dapagliflozin between subjects with normal renal function and patients with type 2 diabetes mellitus who have renal impairment^{a)}

Analyte	Day		Pharmacokinetic parameter	Patients with mild renal impairment	Patients with moderate renal impairment	Patients with severe renal impairment	
Unchanged dapagliflozin	Period I (single-dose administration)		C_{max}	1.142 [1.052, 1.239]	1.256 [1.091, 1.445]	1.355 [1.123, 1.633]	
			AUC_{inf}	1.278 [1.189, 1.374]	1.523 [1.346, 1.724]	1.753 [1.486, 2.068]	
			AUC_{0-t}	1.246 [1.164, 1.334]	1.459 [1.298, 1.639]	1.654 [1.416, 1.933]	
	Period II (multiple-dose administration)	Day 4	C_{max}	1.124 [1.009, 1.252]	1.222 [1.016, 1.469]	1.306 [1.021, 1.671]	
			AUC_{τ}	1.227 [1.080, 1.394]	1.419 [1.140, 1.766]	1.595 [1.191, 2.135]	
		Day 10	C_{max}	1.037 [0.922, 1.167]	1.064 [0.870, 1.302]	1.087 [0.830, 1.422]	
			AUC_{τ}	1.316 [1.137, 1.523]	1.602 [1.247, 2.057]	1.874 [1.342, 2.617]	
Dapagliflozin 3-O-glucuronide	Period I (single-dose administration)		C_{max}	1.199 [1.111, 1.293]	1.364 [1.198, 1.554]	1.513 [1.272, 1.799]	
			AUC_{inf}	1.503 [1.366, 1.654]	2.011 [1.707, 2.369]	2.538 [2.039, 3.158]	
			AUC_{0-t}	1.467 [1.339, 1.608]	1.929 [1.649, 2.257]	2.402 [1.948, 2.961]	
	Period II (multiple-dose administration)	Day 4	C_{max}	1.125 [1.010, 1.253]	1.224 [1.018, 1.471]	1.309 [1.023, 1.673]	
			AUC_{τ}	1.417 [1.255, 1.599]	1.816 [1.477, 2.235]	2.216 [1.681, 2.922]	
		Day 10	C_{max}	1.200 [1.109, 1.298]	1.366 [1.193, 1.564]	1.516 [1.266, 1.816]	
			AUC_{τ}	1.542 [1.350, 1.760]	2.100 [1.673, 2.636]	2.689 [1.986, 3.642]	

Estimated geometric mean ratio (patients with renal impairment/subjects with normal renal function) and its two-sided 90% CI
 C_{max} , Maximum plasma concentration; AUC_{inf} , Area under the plasma concentration-time curve from time 0 to infinity; AUC_{0-t} , Area under the plasma concentration-time curve from 0 to the last sampling point (t); AUC_{τ} , Area under the concentration-time curve at dosing interval at steady state

a) Severity of renal impairment was defined according to the iohexol clearance: subjects with normal renal function (healthy adult subjects, patients with type 2 diabetes mellitus), 100 mL/min; patients with mild renal impairment, 65 mL/min; patients with moderate renal impairment, 40 mL/min; patients with severe renal impairment, 20 mL/min.

The mean plasma protein binding of unchanged dapagliflozin in each group ranged from 92.3% to 94.6%.

Regarding pharmacodynamics, the two-sided 95% CI of the slope of the regression line between renal glucose clearance¹¹⁴ and CLcr during the first 6 hours after administration was positive and did not contain zero. Based on this regression analysis, a comparison of renal glucose clearance during the first 6 hours after dapagliflozin administration between subjects with normal renal function (healthy adult subjects, patients with type 2 diabetes mellitus) and patients with type 2 diabetes mellitus who have renal impairment was performed. The results were as shown in Table 12.

correlation coefficient was 0.86 between iohexol systemic clearance and creatinine clearance calculated by Cockcroft-Gault formula, and 0.76 between iohexol systemic clearance and 24-hour creatinine clearance.

¹¹³ Regression analysis of parameters after multiple oral administration was performed using values on Days 4 and 10.

¹¹⁴ Renal glucose clearance during the first 6 hours after administration = 6-hour cumulative urinary glucose excretion after administration of dapagliflozin/serum glucose AUC_{0-6h}

Table 12. Renal glucose clearance during the first 6 hours after administration in patients with type 2 diabetes mellitus who have renal impairment relative to that in subjects with normal renal function^{a)}

Day	Patients with mild renal impairment	Patients with moderate renal impairment	Patients with severe renal impairment
Day 1	-11.81 [-13.48, -10.15]	-20.25 [-23.11, -17.40]	-27.00 [-30.81, -23.20]
Day 4	-8.37 [-11.98, -4.75]	-14.34 [-20.54, -8.15]	-19.12 [-27.38, -10.86]
Day 10	-14.13 [-17.83, -10.43]	-24.22 [-30.57, -17.87]	-32.30 [-40.77, -23.83]

Difference in estimated means^{b)} (patients with renal impairment - subjects with normal renal function) and its two-sided 95% CI

Renal glucose clearance was calculated as $(0.0666 + 0.3375 \times CLcr)$ for Day 1, $(2.6874 + 0.2390 \times CLcr)$ for Day 4, and $(0.1345 + 0.4037 \times CLcr)$ for Day 10.

a) Subjects with normal renal function on Day 1 were defined as healthy adult subjects and patients with normal renal function, and subjects with normal renal function on Days 4 and 10 were defined as patients with normal renal function.

b) The estimated means were obtained from linear regression analysis between 6-hour renal glucose clearance and 6-hour CLcr on Days 1, 4, and 10.

The 24-hour cumulative urinary glucose excretions (mean \pm SD) after administration of dapagliflozin in patients with normal renal function and in patients with mild, moderate, and severe renal impairment were 84.46 ± 38.79 , 42.80 ± 25.11 , 25.72 ± 15.54 , and 11.99 ± 7.26 g, respectively, on Day 1; 54.39 ± 27.07 , 41.85 ± 33.38 , 18.08 ± 11.62 , and 16.71 ± 11.89 g, respectively, on Day 4; and 84.86 ± 42.56 , 51.83 ± 38.74 , 17.53 ± 10.97 , and 10.73 ± 5.99 g, respectively, on Day 10.

Regarding safety, 26 adverse events were reported by 14 of 40 subjects. Among patients with normal renal function, 2 adverse drug reactions (hypoglycaemia, headache) occurred in 2 of 12 subjects during Period I and 1 adverse drug reaction (headache) in 1 of 4 subjects during Period II. Among patients with mild renal impairment, 5 adverse drug reactions (diarrhoea, nausea/vomiting asthenia/inappetence) occurred in 2 of 4 subjects during Period II. Among patients with severe renal impairment, 1 adverse drug reaction (blood creatinine increased) occurred in 1 of 4 subjects during Period I. No deaths or serious adverse events were reported. Adverse events leading to treatment discontinuation were reported by 2 subjects (exacerbation of gastrooesophageal reflux disease in a patient with severe renal impairment during Period I, dizziness in a patient with moderate renal impairment during Period II), but a causal relationship to the study drug was ruled out for these events. A total of 71 laboratory abnormalities were reported by 36 subjects; the most commonly reported event was increased urine sugar (25 subjects), followed by blood urea nitrogen (BUN) increased (11 subjects). No marked differences were seen in serum electrolytes between before and after administration of dapagliflozin, and no clinically meaningful changes were observed in total urinary protein. No changes of specific concern were seen in vital signs or ECG.

4.(ii).A.(4).2 Study in subjects with hepatic impairment (5.3.3.3.2, Study MB102027 [March to October 2008])

An open-label, parallel-group study was conducted to evaluate the pharmacokinetics and safety of a single oral dose of dapagliflozin in foreign healthy adult subjects and subjects with hepatic impairment (target sample size, 24).

A single oral dose of dapagliflozin 10 mg was to be administered under fasted conditions.

All of the 24 treated subjects (6 subjects in the healthy adult subject group, 6 subjects in the Child-Pugh A group, 6 subjects in the Child-Pugh B group, and 6 subjects in the Child-Pugh C group) were included in the safety and pharmacokinetic analysis sets.

Regarding pharmacokinetics, the pharmacokinetic parameters of unchanged dapagliflozin and dapagliflozin 3-O-glucuronide after single administration of dapagliflozin were as shown in Table

13, and comparison of pharmacokinetic parameters after a single dose of dapagliflozin between subjects with hepatic impairment and healthy adult subjects are shown in Table 14.

Table 13. Pharmacokinetic parameters of unchanged dapagliflozin and dapagliflozin 3-O-glucuronide after single oral administration of dapagliflozin

Analyte	Hepatic impairment	No. of subjects	C _{max} (ng/mL)	t _{max} (h)	AUC _{0-t} (ng·h/mL)	AUC _{inf} (ng·h/mL)	t _{1/2} (h)	CL/F (L/h)	Vz/F (L)
Unchanged dapagliflozin	Healthy adult subjects	6	136 (31)	1.00 (0.50, 2.00)	438 (34)	465 (34)	12.9 ± 5.54	21.5 (35)	370 (36)
	Child-Pugh A	6	120 (28)	1.25 (0.50, 3.17)	443 (25)	480 (26)	15.0 ± 16.26	20.8 (28)	322 (91)
	Child-Pugh B	6	153 (51)	0.75 (0.50, 3.00)	614 (40)	632 (40)	8.1 ± 2.87	15.8 (29)	174 (52)
	Child-Pugh C	6	190 (40)	0.75 (0.50, 4.00)	762 (22)	776 (22)	6.1 ± 1.35	12.9 (23)	111 (28)
Dapagliflozin 3-O-glucuronide	Healthy adult subjects	6	196 (41)	1.25 (1.00, 3.00)	803 (38)	837 (38)	16.4 ± 15.16	-	-
	Child-Pugh A	6	203 (60)	2.00 (1.00, 4.17)	853 (46)	889 (44)	10.5 ± 5.14	-	-
	Child-Pugh B	6	310 (53)	1.75 (1.50, 3.00)	1650 (49)	1670 (49)	9.3 ± 7.24	-	-
	Child-Pugh C	6	168 (43)	2.00 (1.50, 6.00)	1049 (34)	1082 (32)	6.1 ± 1.67	-	-

C_{max}, AUC_{0-t}, AUC_{inf}, CL/F, and Vz/F are expressed in the geometric mean (CV%); t_{max} in median (minimum, maximum); and t_{1/2} in mean ± SD.

C_{max}, Maximum plasma concentration; t_{max}, Time to reach the maximum plasma concentration; AUC_{0-t}, Area under the plasma concentration-time curve from 0 to the last sampling point (t); AUC_{inf}, Area under the plasma concentration-time curve from 0 to infinity; t_{1/2}, Elimination half-life; CL/F, Oral clearance; Vz/F, Apparent volume of distribution; -, Not applicable

Table 14. Comparison of pharmacokinetic parameters after single oral administration of dapagliflozin between subjects with hepatic impairment and healthy adult subjects

Analyte	Pharmacokinetic parameter	Child-Pugh A	Child-Pugh B	Child-Pugh C
Unchanged dapagliflozin	C _{max}	0.882 [0.598, 1.301]	1.122 [0.761, 1.654]	1.395 [0.946, 2.056]
	AUC _{inf}	1.033 [0.765, 1.396]	1.359 [1.007, 1.836]	1.669 [1.236, 2.255]
	AUC _{0-t}	1.011 [0.750, 1.363]	1.401 [1.040, 1.889]	1.739 [1.291, 2.345]
Dapagliflozin 3-O-glucuronide	C _{max}	1.035 [0.651, 1.646]	1.582 [0.995, 2.515]	0.857 [0.539, 1.362]
	AUC _{inf}	1.062 [0.705, 1.600]	1.996 [1.325, 3.007]	1.293 [0.858, 1.947]
	AUC _{0-t}	1.063 [0.699, 1.617]	2.055 [1.351, 3.126]	1.307 [0.859, 1.988]

Estimated geometric mean ratio (subjects with hepatic impairment/healthy adult subjects) and its two-sided 90% CI

C_{max}, Maximum plasma concentration; AUC_{inf}, Area under the plasma concentration-time curve from time 0 to infinity; AUC_{0-t}, Area under the plasma concentration-time curve from 0 to the last sampling point (t)

The geometric mean (CV%) of the molar ratio¹¹⁵ was 1.26 (38), 1.29 (50), 1.85 (46), and 0.97 (17) in the healthy adult subject group, and the Child-Pugh A, B, and C groups, respectively. The mean plasma protein binding of unchanged dapagliflozin in each group of subjects was in the range of 91.1% to 93.4%.

Regarding safety, an adverse event was reported by 1 of 6 subjects (1 event, phlebitis) in the healthy adult subject group, 2 of 6 subjects (3 events; abdominal discomfort/back pain, rash) in the Child-Pugh B group, and 1 of 6 subjects (1 event, dizziness) in the Child-Pugh C group, but a causal relationship to the study drug was ruled out for these events. There were no deaths, serious adverse events, or adverse events leading to treatment discontinuation. A total of 25 laboratory abnormalities were reported by 13 subjects (3 healthy subjects, 1 subject in the Child-Pugh A group, 3 subjects in the Child-Pugh B group, 6 subjects in the Child-Pugh C group). Among these, 18 laboratory abnormalities were reported by subjects in the Child-Pugh C group, and none was considered as clinically significant. No changes of specific concern were seen in

¹¹⁵ Molar ratio = (AUC_{inf} of dapagliflozin 3-O-glucuronide/AUC_{inf} of unchanged dapagliflozin) × (molecular weight of dapagliflozin/molecular weight of dapagliflozin 3-O-glucuronide)

vital signs. Five events of electrocardiogram abnormal were reported by 5 subjects (3 subjects in the Child-Pugh A group, 1 subject in the Child-Pugh B group, 1 subject in the Child-Pugh C group), but none was assessed as an adverse event.

4.(ii).A.(5) Drug-drug interaction studies (5.3.3.4.1 to 5.3.3.4.10; Study MB102004 [September to October 2005], Study MB102017 [June to July 2007], Study MB102026 [November 2007 to February 2008], Study MB102036 [February to March 2009], Study MB102037 [March to May 2009], Study MB102057 [July to September 2009], Study MB102058 [June to August 2009], Study MB102074 [April 2010], Study MB102093 [November 2010], Study D1690C00002 [January to April 2010])

The results of the conducted drug-drug interaction studies were as shown in Table 15. All studies except Study D1690C00002 in Japanese patients with type 2 diabetes mellitus were conducted in foreign healthy adult subjects.

Table 15. Results of drug-drug interaction studies

Study No.	Dose of dapagliflozin	Name and dose of concomitant drug	Analyte	Comparison of plasma pharmacokinetic parameters between monotherapy and concomitant therapy with concomitant drug	
				C _{max}	AUC _{inf}
MB102004 ^{a)}	50 mg	Hydrochlorothiazide 25 mg	Unchanged dapagliflozin (n = 18)	-	1.07 [1.04, 1.11]
			Hydrochlorothiazide (n = 18)	-	0.99 [0.95, 1.04]
MB102017 ^{a)}	50 mg	Pioglitazone 45 mg	Unchanged dapagliflozin (n = 24)	1.09 [1.00, 1.18]	1.03 [0.98, 1.08]
			Pioglitazone (n = 24)	0.93 [0.75, 1.15]	1.00 [0.90, 1.13]
			Hydroxioglitzzone ^{b)} (n = 24)	0.90 [0.79, 1.02]	1.05 [0.90, 1.22]
MB102026 ^{a)}	20 mg	Metformin 1000 mg	Unchanged dapagliflozin (n = 18)	0.932 [0.848, 1.024]	0.995 [0.945, 1.053]
			Metformin (n = 18)	0.953 [0.866, 1.049]	1.001 [0.933, 1.075]
MB102036 ^{b)}	20 mg	Simvastatin 40 mg	Unchanged dapagliflozin (n = 24)	0.978 [0.887, 1.078]	0.986 [0.957, 1.017]
			Simvastatin (n = 24)	0.936 [0.816, 1.073]	1.193 [1.018, 1.399]
			Simvastatin acid ^{d)} (n = 24)	1.077 [0.931, 1.247]	1.311 [1.146, 1.499]
		Valsartan 320 mg	Unchanged dapagliflozin (n = 24)	0.881 [0.796, 0.975]	1.024 [1.000, 1.049]
			Valsartan (n = 24)	0.938 [0.762, 1.156]	1.046 [0.850, 1.286]
MB102037 ^{b)}	20 mg	Glimepiride 4 mg	Unchanged dapagliflozin (n = 18)	1.006 [0.921, 1.097]	0.989 [0.958, 1.020]
			Glimepiride (n = 18)	1.043 [0.905, 1.201]	1.132 [0.996, 1.287]
		Sitagliptin 100 mg	Unchanged dapagliflozin (n = 18)	0.958 [0.875, 1.049]	1.081 [1.031, 1.133]
			Sitagliptin (n = 18)	0.887 [0.807, 0.974]	1.012 [0.985, 1.040]
MB102057 ^{c)}	10 mg	Bumetanide 1 mg	Unchanged dapagliflozin (n = 42)	1.132 [0.979, 1.310]	1.132 ^{j)} [0.985, 1.302]
			Bumetanide (n = 42)	1.080 [0.953, 1.222]	1.047 ^{j)} [0.991, 1.106]
MB102058 ^{d)}	10 mg	Warfarin 25 mg	Unchanged dapagliflozin	-	-
			S-warfarin (n = 14)	1.030 [0.994, 1.124]	1.068 [1.002, 1.138]
	10 mg	Digoxin 0.25 mg	R-warfarin (n = 14)	1.057 [0.977, 1.145]	1.079 [1.030, 1.130]
			Unchanged dapagliflozin	-	-
			Digoxin (n = 16)	0.990 [0.843, 1.162]	1.002 ^{k)} [0.860, 1.167]
MB102074 ^{e)}	10 mg	Rifampicin 600 mg	Unchanged dapagliflozin (n = 14)	0.931 [0.779, 1.112]	0.780 [0.731, 0.832]
MB102093 ^{f)}	10 mg	Mefenamic acid 250 mg	Unchanged dapagliflozin (n = 16)	1.13 [1.03, 1.24]	1.51 [1.44, 1.58]
D1690C00002 ^{g)}	10 mg	Voglibose 0.2 mg	Unchanged dapagliflozin (n = 22)	1.040 [0.899, 1.204]	1.009 [0.954, 1.067]

Geometric mean ratios (concomitant therapy with concomitant drug/monotherapy) and their two-sided 90% CI of plasma pharmacokinetic parameters of unchanged dapagliflozin and the concomitant drug; -, Not calculated.

a) Three-period crossover study: Both drugs were administered as a single oral dose after fasting for ≥10 hours.

b) Five-period crossover study: Both drugs were administered as a single oral dose after fasting for ≥10 hours.

- c) Subjects in the bumetanide group received 1 mg of bumetanide alone from Day 1 to Day 7 and received bumetanide 1 mg + dapagliflozin 10 mg from Day 8 to Day 14. Subjects in the dapagliflozin group received dapagliflozin 10 mg alone from Day 1 to Day 7 and received bumetanide 1 mg + dapagliflozin 10 mg from Day 8 to Day 14. Subjects in the concomitant therapy group received bumetanide 1 mg + dapagliflozin 10 mg from Day 1 to Day 14. (Both drugs were orally administered once daily at 1 hour after breakfast.)
- d) Two-group, two-treatment, two-period crossover study: Subjects in the warfarin concomitant therapy group received dapagliflozin 20 mg alone on Day 1, dapagliflozin 10 mg + warfarin 25 mg on Day 2, and dapagliflozin 10 mg alone from Day 3 to Day 8 (both drugs were orally administered once daily under fasted conditions), and the warfarin monotherapy group received a single oral dose of warfarin 25 mg alone on Day 2 under fasted conditions. Subjects in the digoxin concomitant therapy group received dapagliflozin 20 mg alone on Day 1, dapagliflozin 10 mg + digoxin 0.25 mg on Day 2, and dapagliflozin 10 mg alone from Day 3 to Day 8 (both drugs were orally administered once daily under fasted conditions), and the digoxin monotherapy group received a single oral dose of digoxin 0.25 mg alone on Day 2 under fasted conditions.
- e) Subjects received dapagliflozin 10 mg alone on Day 1, rifampicin 600 mg alone on Day 4 to Day 8, dapagliflozin 10 mg + of rifampicin 600 mg on Day 9, and rifampicin 600 mg alone on Day 10 to Day 11 (both drugs were orally administered once daily under fasted conditions).
- f) Subjects received a single oral dose of dapagliflozin 10 mg under fasted conditions on Day 1, an oral dose of mefenamic acid 500 mg under fed conditions followed by 3 oral doses of mefenamic acid 250 mg with a meal with a 6-hour interval between doses on Day 4, and a single oral dose of dapagliflozin 10 mg + mefenamic acid 250 mg under fasted conditions followed by 11 oral doses of mefenamic acid 250 mg with a meal with a 6-hour interval between doses on Day 5 to Day 8.
- g) An open-label study: Subjects received dapagliflozin 10 mg (a single oral dose) + voglibose 0.2 mg (orally administered 3 times daily immediately before a meal) in Period I, and dapagliflozin 10 mg alone (a single oral dose) in Period II.
- h) An active metabolite of pioglitazone
- i) An active metabolite of simvastatin
- j) Ratio of AUC_{τ}
- k) Ratio of AUC_{0-t}

Regarding pharmacodynamics, in the drug-drug interaction study with hydrochlorothiazide (Study MB102004), urinary Na excretion was increased from baseline by approximately 32 mEq/24 h with dapagliflozin monotherapy, by approximately 60 mEq/24 h with hydrochlorothiazide monotherapy, and by approximately 126 mEq/24 h with concomitant therapy with these 2 drugs. No apparent differences were observed in urinary glucose excretion between dapagliflozin monotherapy and the concomitant therapy.

In the drug-drug interaction study with bumetanide (Study MB102057), urinary glucose excretion was observed during monotherapy in the dapagliflozin group (approximately 30 g/24 h on Day 7), but not during monotherapy in the bumetanide group.

In all groups, urine output was increased from baseline by approximately 800 to 1200 mL/24 h on Day 1, but decreased thereafter. During concomitant therapy of dapagliflozin with bumetanide on Day 8 in both the bumetanide and dapagliflozin groups, urine output was increased by approximately 700 to 1000 mL/24 h compared with that after monotherapy with each drug on Day 7, but decreased thereafter.

Urinary Na excretion increased until 1.5 hours post-dose during bumetanide monotherapy, but decreased thereafter to near baseline values within 3.5 to 4 hours post-dose; a similar trend was observed during steady state in the concomitant therapy group and on Day 8 in the dapagliflozin group (the first day of the concomitant therapy). Urinary Na excretion increased from baseline by approximately 80 mEq/24 h on Day 1 in the bumetanide group (during bumetanide monotherapy) and in the concomitant therapy group, and then decreased to near baseline. In the dapagliflozin group, urinary Na excretion increased from baseline by approximately 20 mEq/24 h on Day 1 (during dapagliflozin monotherapy) and then remained at the same levels. On Day 8 (during concomitant use of dapagliflozin with bumetanide), there was a transient increase in urinary Na excretion in the dapagliflozin and bumetanide groups (by approximately 100 mEq/24 h in the dapagliflozin group and approximately 60 mEq/24 h in the bumetanide group compared with Day 7 [during monotherapy with each drug]).

Urine osmolarity did not substantially change during monotherapy in the bumetanide group, but increased from baseline by approximately 60 to 170 mOsm/kg on each day during concomitant

therapy in the dapagliflozin and bumetanide groups. In the concomitant therapy group, however, urine osmolarity increased from base line by approximately 50 to 90 mOsm/kg on each day.

Urinary uric acid excretion increased transiently during treatment with dapagliflozin, but then gradually declined to near baseline. Urinary uric acid excretion decreased during monotherapy in the bumetanide group. Serum uric acid concentration was decreased from baseline by 1.94 mg/dL during monotherapy on Day 8 in the dapagliflozin group, and by 1.24 mg/dL on Day 8 in the concomitant therapy group.¹¹⁶ Serum uric acid concentration in the bumetanide group was similar to baseline until Day 8 (the starting day of bumetanide monotherapy), and was decreased on Day 15 (the seventh day of concomitant use of bumetanide and dapagliflozin) by 1.62 mg/dL compared with that on Day 8 (baseline for concomitant use of dapagliflozin with bumetanide).

Plasma renin activity did not change during dapagliflozin monotherapy in the dapagliflozin group, while it slightly increased (by approximately 0.5-4 ng/mL·h) from baseline during bumetanide monotherapy and concomitant therapy of dapagliflozin with bumetanide in the bumetanide group.

No clinically meaningful changes were observed in other urine electrolytes (K, chloride [Cl], calcium [Ca], magnesium [Mg], phosphorus [P], creatinine) or clinical chemistry (Na, K, Cl, Ca, Mg, P, bicarbonate, creatinine, BUN, osmolarity).

In the drug-drug interaction study with warfarin and digoxin (Study MB102058), the adjusted geometric mean ratio [two-sided 90% CI] (concomitant therapy/warfarin monotherapy) for INR_{max} and AUC_{INR} was 1.004 [0.967, 1.043] and 1.007 [0.989, 1.025], respectively.

In the drug-drug interaction study with rifampicin (Study MB102074), the least-squares mean difference [two-sided 90% CI] (concomitant use with rifampicin - dapagliflozin monotherapy) in 24-hour cumulative urinary glucose excretion was -5171 [-9166, -1177] mg (creatinine-corrected value, -4.67 [-6.77, -2.57] mg/mg Cr).

In the drug-drug interaction study with mefenamic acid (Study MB102093), the cumulative urinary glucose excretions (mean ± SD) from 0 to 24, 24 to 48, and 48 to 72 hours post-dose were 43.5 ± 9.17, 18.9 ± 7.95, and 5.4 ± 4.67 g, respectively, during dapagliflozin monotherapy and 51.5 ± 8.31, 31.4 ± 9.24, and 10.6 ± 6.67 g, respectively, during concomitant therapy. The least-squares mean difference [two-sided 95% CI] (concomitant therapy - dapagliflozin monotherapy) in 24-hour cumulative urinary glucose excretion was 7.97 [3.17, 12.77] g.

4.(ii).A.(6) Pharmacodynamic studies

Thorough QT/QTc study (5.3.4.1.1, Study D1690C00001 [July 2007 to April 2008])

A randomized, double-blind, four-period, placebo- and moxifloxacin- (positive control) controlled crossover study was conducted to evaluate the effects of a single oral dose of dapagliflozin on QTc interval in foreign healthy adult male subjects (target sample size, 36).

¹¹⁶ The applicant discussed that the mechanism of the decrease in serum uric acid levels by dapagliflozin may be explained by promotion of urinary glucose excretion resulting from dapagliflozin administration and from decreased renal uric acid absorption by SLC2A9a caused by high glucose levels in the renal tubules, based on the following reports: In patients with familial renal glycosuria, renal uric acid excretion has been suggested to be enhanced by high concentrations of glucose in the proximal renal tubules (Skeith MD, et al. *Am J Physiol*. 1970;219:1080-2); In a rodent study using a model of perfused renal tubules, reabsorption of uric acid has been reported to be inhibited by high concentrations of glucose in the renal tubules, without being affected by phlorizin, a nonspecific SGLT inhibitor (Knight TF, et al. *Am J Physiol*. 1979;236:526-9); and renal uric acid transport via SLC2A9a (glucose/uric acid exchanger) has been reported to be balanced toward an elimination predominant state by high concentrations of glucose in the renal tubules (Caulfeld MJ, et al. *PLoS Medicine*. 2008;5:1509-23, Witkowska K, et al. *Am J Physiol*. 2012;303:527-39).

A single oral dose of placebo, dapagliflozin 20 or 150 mg, or moxifloxacin 400 mg was to be administered under fasted conditions. A washout period of 7 to 10 days was required between each period.

All of the 50 treated subjects were included in the safety analysis set, 47 subjects¹¹⁷ were included in the pharmacokinetic analysis sets and 36 subjects¹¹⁸ were included in the pharmacodynamic analysis sets.

A total of 13 subjects discontinued the study, and the reasons for study discontinuation included consent withdrawal (6 subjects), non-compliance with the study protocol (3 subjects), safety reasons (2 subjects), an adverse event (1 subject), and others (1 subject).

Regarding pharmacokinetics, the geometric means (% coefficient of variation) of C_{max} of dapagliflozin after single oral administration of dapagliflozin at doses of 20 and 150 mg were 250.7 (26.6) and 1981.0 (37.8) ng/mL, respectively, and the median t_{max} (minimum, maximum) were 1 (0.5, 3) and 1 (0.5, 4) h, respectively.

Regarding ECG, the difference in the mean change (least squares mean) [two-sided 90% CI] in QTcX interval¹¹⁹ from baseline between the dapagliflozin and placebo groups ($\Delta\Delta QTcX$) peaked (2.3 [0.0, 4.6] msec) at 8 hours after administration of dapagliflozin 20 mg, while peaked (1.2 [-1.0, 3.4] msec) at 3 hours after administration of dapagliflozin 150 mg, the upper limit of the CI was lower than 10 msec at both dose levels. However, after administration of moxifloxacin, $\Delta\Delta QTcX$ peaked (9.7 [7.5, 11.9] msec) at 4 hours post-dose, and the estimated $\Delta\Delta QTcX^{120}$ [two-sided 90% CI] at 1, 2, 3, and 4 hours post-dose was 7.7 [6.2, 9.1] msec, the lower limit of the CI was higher than 5 msec.

Regarding safety, 5 adverse events occurred in 5 of 46 subjects receiving placebo, 7 adverse events in 5 of 41 subjects receiving dapagliflozin 20 mg, 8 adverse events in 5 of 44 subjects receiving dapagliflozin 150 mg, and 7 adverse events in 5 of 41 subjects receiving moxifloxacin. Four adverse drug reactions (palpitations, headache, pruritus/urticaria) occurred in 3 of 41 subjects receiving dapagliflozin 20 mg, 2 adverse drug reactions (2 events of headache) in 2 of 44 subjects receiving dapagliflozin 150 mg, and 1 adverse drug reaction (dyspnoea) in 1 of 41 subjects receiving moxifloxacin. Adverse events leading to treatment discontinuation were reported by 1 subject receiving dapagliflozin 150 mg (headache/myalgia/pharyngolaryngeal pain), but a causal relationship to the study drug was ruled out for these events. No deaths or serious adverse events were reported.

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

4.(ii).B.(1) PK/PD relationship

The applicant explained the PK/PD relationship of dapagliflozin as follows:

A relationship between the dose level and pharmacodynamic effects (change from baseline in 24-hour urinary glucose excretion) of dapagliflozin was investigated based on the E_{max} model which included subjects' health status (healthy adult subjects or patients with type 2 diabetes mellitus) as a covariate. As a result, the dose level that produces 50% of the maximal effect was 4.70 mg

¹¹⁷ Two subjects who discontinued the study on their own decision and 1 subject who was non-compliant with the study protocol were excluded from the pharmacokinetic analysis set.

¹¹⁸ The following subjects were excluded from the pharmacodynamic analysis set: 6 subjects who discontinued the study on their own decision; 1 subject with missing PD data at baseline; 3 subjects who were non-compliant with the study protocol; 2 subjects who discontinued the study due to safety reasons; 1 subject who discontinued the study due to an adverse event; and 1 subject who discontinued the study due to physician's decision.

¹¹⁹ QT interval corrected for heart rate using a study-specific correction factor

¹²⁰ Analysis was performed using repeated measures ANCOVA model with subject, study drug, period, time, interaction between period and time, and interaction between the study drug and time as independent factors and baseline QTcX interval as a covariate. Subjects within sequences were included as a random effect.

in healthy adult subjects and 3.97 mg in patients with type 2 diabetes mellitus, and the maximum change from baseline in urinary glucose excretion during the first 24 hours post-dose was higher in patients with type 2 diabetes mellitus than in healthy adult subjects, which was estimated to be 66.2 g in healthy adult subjects and 83.1 g in patients with type 2 diabetes mellitus. In addition, dapagliflozin was suggested to produce its maximal effect at a dose of 10 mg.

Since a relationship between C_{max} and AUC of unchanged dapagliflozin and the change from baseline in 24-hour urinary glucose excretion was found to be similar to that between the dose level and the change from baseline in 24-hour urinary glucose excretion, the former relationship was evaluated using the E_{max} model in the same manner. As a result, the estimated C_{max} that produces 50% of the maximal effect was 50.92 ng/mL in healthy adult subjects and 53.92 ng/mL in patients with type 2 diabetes mellitus, and the estimated maximum change from baseline in urinary glucose excretion during the first 24 hours post-dose was 65.95 g in healthy adult subjects and 82.72 g in patients with type 2 diabetes mellitus. Similarly, the estimated AUC that produces 50% of the maximal effect was 198.3 ng·h/mL in healthy adult subjects and 197.9 ng·h/mL in patients with type 2 diabetes mellitus, and the estimated maximum change from baseline in urinary glucose excretion during the first 24 hours post-dose was 63.70 g in healthy adult subjects and 80.53 g in patients with type 2 diabetes mellitus.

A relationship between the dose level of dapagliflozin and the renal glucose clearance during the first 4 or 6 hours post-dose in healthy adult subjects and patients with type 2 diabetes mellitus¹²¹ was investigated based on the E_{max} model. As a result, the estimated dose level that produces 50% of the maximal effect was 0.98 mg and the estimated highest renal glucose clearance was 33.81 mL/min. In a similar evaluation of relationship between C_{max} and AUC of unchanged dapagliflozin and the renal glucose clearance, the estimated C_{max} that produces 50% of the maximal effect was 13.02 ng/mL and the estimated highest renal glucose clearance was 33.81 mL/min, and the estimated AUC that produces 50% of the maximal effect was 46.70 ng·h/mL and the estimated highest renal glucose clearance was 33.39 mL/min.

PMDA asked the applicant to explain the effects of age and sex on the exposure of dapagliflozin and urinary glucose excretion.

The applicant responded as follows:

Regarding the effects of age, the exposure in the elderly subjects (≥ 65 years of age) was estimated to be higher by approximately 25% than that in the control subjects (≥ 40 and < 65 years of age) based on a pooled population pharmacokinetic analysis¹²² in foreign subjects. The difference in exposure is considered attributable to renal impairment because renal impairment is commonly observed in the elderly. The age is considered to have little effect on the urinary glucose excretion induced by dapagliflozin, because a residual plot based on the above mentioned E_{max} model evaluating the relationship between age and the change in urinary glucose excretion in the elderly subjects showed no age-related bias.

As for the effect of sex, based on a pooled analysis of clinical pharmacology studies, exposure in female subjects was estimated to be higher than that in male subjects by about 23.4% at most. In a population pharmacokinetic analysis in foreign subjects¹²² and in Japanese subjects (Studies MB102025 and D1692C00005), sex was estimated as a covariate for CL/F. However, a residual plot based on the above mentioned E_{max} model evaluating the relationship between sex and the

¹²¹ Because the renal glucose clearance enhancing effect of dapagliflozin was similar between healthy adult subjects and patients with type 2 diabetes mellitus, subjects' health status (healthy adult subjects or patients with type 2 diabetes mellitus) was not included in the analysis as a covariate.

¹²² Based on the results of population pharmacokinetic analysis using 8011 measured values of plasma parent concentration of dapagliflozin obtained from 30 healthy adult subjects and 1223 patients with type 2 diabetes mellitus in foreign clinical studies (Studies MB102002, MB102003, MB102013, MB102032, and D1690C00006). CLcr and sex were estimated as covariates for CL/F, and body weight was estimated as a covariate on V2/F.

change in urinary glucose excretion showed no gender-related differences. In addition, no gender-related differences were observed in the change in HbA1c during long-term treatment. Based on the above, any difference in exposure according to age or sex would not result in a substantial difference in the urinary glucose excretion.

PMDA accepted the applicant's response. However, the use of dapagliflozin in the elderly patients will be additionally reviewed from a safety standpoint in the clinical section [see "4.(iii).B.(6).3) Elderly patients"].

4.(ii).B.(2) Pharmacokinetics in patients with renal or hepatic impairment

Since the exposure to dapagliflozin increased in patients with renal or hepatic impairment depending on its severity, PMDA asked the applicant to explain the safety in these patient populations.

The applicant responded as follows:

The effect of renal function on the pharmacokinetics of dapagliflozin was evaluated in patients with mild, moderate, and severe renal impairment and patients with type 2 diabetes mellitus who have normal renal function (Study MB102007). Following multiple oral administration of dapagliflozin 20 mg once daily for 7 days, the estimated geometric means of C_{max} and AUC_{τ} of unchanged dapagliflozin on Day 7 in patients with mild renal impairment increased by 4% and 32%, respectively, those in patients with moderate renal impairment increased by 6% and 60%, respectively, and those in patients with severe renal impairment increased by 9% and 87%, respectively, compared with patients with type 2 diabetes mellitus who had normal renal function. The exposure and eGFR data from the foreign phase III study (Study MB102029) in patients with moderate renal impairment were fit to a regression model to estimate exposure after administration of dapagliflozin 10 mg in Japanese patients with type 2 diabetes mellitus who have moderate renal impairment. The results showed that the geometric mean [two-sided 90% CI] of the ratio of AUC_{τ} (patients with the center value of the eGFR range classified in moderate renal impairment [$45 \text{ mL/min}/1.73 \text{ m}^2$]/patients with normal renal function) was [redacted] [redacted, redacted]; therefore, AUC_{τ} in Japanese patients with type 2 diabetes mellitus who have moderate renal impairment was estimated to be approximately [redacted] ng·h/mL. Assuming that this exposure-eGFR relationship can be extrapolated beyond the eGFR range observed in Foreign Study MB102029, this exposure value corresponded to that in Japanese patients with type 2 diabetes mellitus who have normal renal function and who receive approximately [redacted] mg of dapagliflozin. Similarly, the ratio of exposure (Japanese patients with type 2 diabetes mellitus who have severe renal impairment/patients with normal renal function) after administration of dapagliflozin 10 mg was estimated to be [redacted] [redacted, redacted] and the estimated AUC_{τ} was approximately [redacted] ng·h/mL, which corresponded to that in Japanese patients with type 2 diabetes mellitus who have normal renal function receiving approximately [redacted] mg of dapagliflozin.

The effect of hepatic function on the pharmacokinetics of dapagliflozin was evaluated in patients with mild, moderate, and severe hepatic impairment (Child Pugh class A, B, and C, respectively) and healthy adult subjects (Study MB102027). Following a single oral dose of dapagliflozin 10 mg, no difference was observed in the protein bindings of dapagliflozin among subjects with hepatic impairment or between subjects with hepatic impairment and healthy adult subjects, and the geometric means of C_{max} and AUC_{inf} of unchanged dapagliflozin in subjects with moderate hepatic impairment increased by 12% and 36%, respectively, and those in subjects with severe hepatic impairment increased by 40% and 67%, respectively, compared with those in healthy adult subjects.

Dapagliflozin is mainly metabolized by UGT1A9 to an inactive metabolite dapagliflozin 3-*O*-glucuronide. UGT1A9 is primarily expressed in the human kidney,¹²³ while dapagliflozin 3-*O*-glucuronide was found to be produced in both the kidney and liver based on the results of an *in vitro* study of dapagliflozin (5.3.2.2.1). In general, contribution of the kidney in metabolic clearance is considered to be smaller than that of the liver, but the intrinsic substrate clearance via glucuronidation in the human kidney observed in an *in vitro* study has been reported to be comparable to or more extensive than that by the human liver,¹²⁴ and in addition, UGT in the kidney has been reported to be indispensable to the systemic and renal metabolic clearances of some compounds.¹²⁵ Because relative contributions of the kidney and liver to the glucuronidation of dapagliflozin depend upon the exposure to dapagliflozin and relative expression level of UGT1A9 in these organs, given the high formation rate of dapagliflozin 3-*O*-glucuronide in the kidney and the relative exposure to dapagliflozin in these organs (including blood flow¹²⁶), both the liver and kidneys are considered to make a large contribution to the metabolism of dapagliflozin, consistent with the findings of increased exposure both in subjects with severe renal and hepatic impairments.

Dapagliflozin posed no major safety problems when used as monotherapy or concomitant therapy with other antidiabetic drugs in patients with type 2 diabetes mellitus, and the observed events such as genital infection, urinary tract infection, and pollakiuria were predictable based on pharmacological activity of dapagliflozin. Adverse events more commonly reported in the dapagliflozin 10 mg group than in the placebo group in a Japanese phase III study (Study D1692C00006) include nasopharyngitis, dental caries, pollakiuria, renal impairment, and back pain, and the events of pollakiuria, renal impairment, and back pain were assessed as adverse drug reactions. In the 10 mg group, the incidence of adverse events in subjects with baseline eGFR of ≥ 30 and < 60 (76.0%) was higher than that in subjects with baseline eGFR of ≥ 60 and < 90 (60.7%), but in general, the incidences of adverse events and adverse drug reactions were not correlated with baseline eGFR or the dose level of dapagliflozin. In addition, dapagliflozin posed no major safety problems when the dose was increased to 10 mg in subjects who were not adequately responsive to dapagliflozin 5 mg in Study D1692C00012.

To evaluate the safety of administration of dapagliflozin 10 mg in patients with moderate or severe renal impairment, the safety data were reviewed for Foreign Study MB102008¹²⁷ in which subjects received multiple administration of dapagliflozin at doses of up to 50 mg for 12 weeks, which represents a dose that yields a higher exposure than that achieved by once daily administration of dapagliflozin 10 mg in patients with renal impairment of moderate or greater severity. The results revealed that events of urinary tract infection, nausea, dizziness, headache, fatigue, back pain, and nasopharyngitis were commonly reported during the double-blind period. The incidence of genitourinary tract infection events during the double-blind period was higher in the high dose groups (20, 50 mg; 16.1%-16.9%) than in the low dose groups (2.5, 5, 10 mg; 6.8%-10.6%), but in general, there was no relationship between adverse events and the dose level.

Based on the above, given the safety results from the Japanese phase II and phase III studies, there were no major problem in patients with renal impairment with eGFR ≥ 45 , but the use of dapagliflozin in patients with renal impairment with eGFR < 45 or end stage renal disease is not recommended because efficacy in these patients cannot be expected. The safety and efficacy in

¹²³ Nishimura M, Naito S. *Drug Metab Pharmacokinet*. 2006;21:357-74

¹²⁴ Tsoutsikos P, et al. *Biochem Pharmacol*. 2004;67:191-9. Bowalgaha K, Miners JO. *Br J Clin Pharmacol*. 2001;52:605-9.

¹²⁵ Knights KM, Miners JO. *Drug Metabol Rev*. 2010;42:63-73.

¹²⁶ Approximately 26% of the cardiac output flows through the liver, and approximately 19.5% of the cardiac output flows through the kidneys (Williams LR, Leggett RM. *Clin Physiol Meas*. 1989;10:187-217.).

¹²⁷ Foreign Study MB102008: A phase II placebo-controlled, randomized, double-blind, parallel-group study in type 2 diabetes mellitus patients without a history of antidiabetic medication who did not have signs of chronic renal insufficiency and who had inadequate glycaemic control with diet and exercise to evaluate the efficacy and safety of monotherapy with dapagliflozin (treatment duration was 12 weeks)

patients with severe hepatic impairment have not been studied, but a statement that the dose may be increased to 10 mg once daily for the patient with an inadequate response to the 5 mg dose if the patient is followed up carefully was included in the proposed package insert. The eligibility and dose level would be determined according to the patient's conditions.

PMDA considers as follows:

Given the fact that the exposure increased with increasing severity of renal impairment, and considering the mechanism of action of dapagliflozin, appropriate caution should be advised in patients with renal impairment. The exposure (AUC) increases by approximately 60% in patients with severe hepatic impairment, and the safety and efficacy in patients with severe hepatic impairment have not been studied. Thus, careful administration of dapagliflozin should be recommended in these patients. The safety in patients with renal or hepatic impairment will be additionally reviewed in the clinical section [see “4.(iii).B.(6).1) Patients with renal impairment” for patients with renal impairment; see “4.(iii).B.(6).2) Patients with hepatic impairment” for patients with hepatic impairment].

4.(iii) Summary of clinical efficacy and safety

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

As the evaluation data, the results from the following studies were submitted: Japanese phase I studies (Studies MB102010, MB102025, and D1692C00002); foreign phase I studies (Studies MB102006, MB102059, D1690C00001, MB102007, MB102027, MB102004, MB102017, MB102026, MB102036, MB102037, MB102057, MB102058, MB102074, and MB102093); Japanese phase II study (Study D1692C00005); and Japanese phase III studies (Studies D1692C00006 and D1692C00012). Primary study results are shown below. HbA1c values are expressed as NGSP values.

4.(iii).A.(1) Clinical pharmacology studies

For a summary and the safety results of main Japanese and foreign phase I studies, see “4.(i) Summary of biopharmaceutic studies and associated analytical methods” and “4.(ii) Summary of clinical pharmacology studies.”

4.(iii).A.(2) Japanese phase II study (5.3.5.1.1, Study D1692C00005 [August 2009 to May 2010])

A placebo-controlled, randomized, double-blind, parallel-group study was conducted to evaluate the efficacy and safety of dapagliflozin in Japanese patients with type 2 diabetes mellitus¹²⁸ (target sample size of 275, 55 subjects per group).

This study consisted of the washout period (6 weeks), single-blind placebo run-in period (4 weeks), double-blind treatment period (12 weeks), and follow-up period (4 weeks).

Placebo or dapagliflozin at a dose of 1, 2.5, 5, or 10 mg were to be orally administered once daily for 12 weeks in the morning.

¹²⁸ Key Inclusion Criteria were patients with type 2 diabetes mellitus aged ≥18 and ≤79 years with fasting C-peptide >1.0 ng/mL and BMI ≤40 kg/m² who met one of the following criteria: 1) Patients without a history of antidiabetic medication at study entry and with HbA1c of ≥7% and ≤10% at study entry; 2) Patients with a limited history of antidiabetic medication before study entry (who received treatment for <30 days from diagnosis; did not receive oral hypoglycemic agents for ≥3 consecutive days or for a total of ≥7 days within 30 days prior to study entry, and received no insulin therapy within 2 weeks prior to study entry) and with HbA1c of ≥7% and ≤10% at study entry (washout not required); 3) Patients with a history of antidiabetic medication before study entry (but not within 6 weeks prior to study entry) and with HbA1c of ≥7% and ≤10% at study entry (washout not required); or 4) Patients with a current history of antidiabetic medication (with one oral hypoglycemic agent or with a combination with 2 types of oral hypoglycemic agents each at a dose less than half of the maximum approved dose) at study entry and with HbA1c of ≤8% and fasting blood glucose of ≤240 mg/dL at study entry (washout required).

All of the treated 279 subjects (54 subjects in the placebo group, 59 subjects in the dapagliflozin 1 mg group, 56 subjects in the dapagliflozin 2.5 mg group, 58 subjects in the dapagliflozin 5 mg group, and 52 subjects in the dapagliflozin 10 mg group) were included in the safety analysis set and full analysis set (FAS). FAS was used for the primary efficacy analysis. A total of 21 subjects discontinued the study during the double-blind treatment period, including 9 subjects in the placebo group (5 subjects due to ineligibility, 1 subject each due to consent withdrawal and safety reasons, 2 subjects due to other reasons), 5 subjects in the dapagliflozin 1 mg group (2 subjects due to ineligibility, 1 subject each due to inclusion/exclusion criteria violation, consent withdrawal, and death), 5 subjects in the dapagliflozin 2.5 mg group (2 subjects each due to ineligibility and consent withdrawal, 1 subject due to an adverse event), and 2 subjects in the dapagliflozin 5 mg group (1 subject each due to an adverse event and consent withdrawal).

The primary efficacy endpoint of the change in HbA1c from baseline (start of the double-blind treatment period) to Week 12 of treatment in the FAS was as shown in Table 16. A statistically significant decrease was observed in all dapagliflozin groups compared with the placebo group ($P < 0.0001$ for all comparisons; ANCOVA model at significance level of 1.5% [two-sided] for each comparison [Dunnett-adjusted significance level, overall significance level for the study was 5% (two-sided)]).

Table 16. Change in HbA1c from baseline to Week 12 of treatment (Study D1692C00005, FAS)

	Placebo group (n = 54)	Dapagliflozin 1 mg group (n = 59)	Dapagliflozin 2.5 mg group (n = 56)	Dapagliflozin 5 mg group (n = 58)	Dapagliflozin 10 mg group (n = 52)
Baseline	8.12 ± 0.714	8.10 ± 0.785	7.92 ± 0.740	8.05 ± 0.660	8.18 ± 0.690
Week 12 ^{a)}	8.48 ± 0.897	7.97 ± 0.883	7.84 ± 0.776	7.68 ± 0.567	7.72 ± 0.703
Change from baseline ^{b)}	0.37 [0.23, 0.50]	-0.12 [-0.25, 0.01]	-0.11 [-0.25, 0.02]	-0.37 [-0.50, -0.24]	-0.44 [-0.58, -0.30]
Difference from the placebo group ^{b)}	-	-0.49 [-0.68, -0.29]	-0.48 [-0.67, -0.28]	-0.74 [-0.93, -0.54]	-0.80 [-1.00, -0.61]
<i>P</i> -value ^{b)c)}	-	<0.0001	<0.0001	<0.0001	<0.0001

Unit, %; mean ± SD; adjusted mean [two-sided 95% CI]; -, Not applicable

a) Last Observation Carried Forward (LOCF)

b) ANCOVA model with treatment as a fixed effect and baseline HbA1c as a covariate

c) Multiplicity of statistical tests was adjusted by using Dunnett's method, and a significance level of 0.015 was used.

The secondary endpoint of the change in fasting blood glucose from baseline to Week 12 of treatment (LOCF, adjusted mean [two-sided 95% CI]) was 11.17 [4.41, 17.93] mg/dL in the placebo group, -15.61 [-22.37, -8.85] mg/dL in the dapagliflozin 1 mg group, -19.83 [-26.46, -13.20] mg/dL in the dapagliflozin 2.5 mg group, -23.51 [-30.27, -16.75] mg/dL in the dapagliflozin 5 mg group, and -31.94 [-38.98, -24.90] mg/dL in the dapagliflozin 10 mg group. The change in body weight from baseline to Week 12 of treatment (LOCF, adjusted mean [two-sided 95% CI]) was -0.05 [-0.42, 0.31] kg in the placebo group, -1.25 [-1.60, -0.90] kg in the dapagliflozin 1 mg group, -1.24 [-1.60, -0.88] kg in the dapagliflozin 2.5 mg group, -2.06 [-2.41, -1.71] kg in the dapagliflozin 5 mg group, and -1.91 [-2.29, -1.54] kg in the dapagliflozin 10 mg group.

Regarding safety, the incidences of adverse events and adverse drug reactions were 38.9% (21 of 54 subjects) and 1.9% (1 of 54 subjects), respectively, in the placebo group; 40.7% (24 of 59 subjects) and 3.4% (2 of 59 subjects), respectively, in the dapagliflozin 1 mg group; 46.4% (26 of 56 subjects) and 1.8% (1 of 56 subjects), respectively, in the dapagliflozin 2.5 mg group; 41.4% (24 of 58 subjects) and 0.0% (0 of 58 subjects), respectively, in the dapagliflozin 5 mg group; and 53.8% (28 of 52 subjects) and 5.8% (3 of 52 subjects), respectively, in the dapagliflozin 10 mg group. Adverse events and adverse drug reactions reported by ≥3% of subjects in any treatment group were as shown in Table 17.

Table 17. Adverse events and adverse drug reactions reported by ≥3% of subjects in any treatment group (Study D1692C00005, safety analysis set)

Event term	Placebo group (n = 54)		Dapagliflozin 1 mg group (n = 59)		Dapagliflozin 2.5 mg group (n = 56)		Dapagliflozin 5 mg group (n = 58)		Dapagliflozin 10 mg group (n = 52)	
	Adverse event	Adverse drug reaction	Adverse event	Adverse drug reaction	Adverse event	Adverse drug reaction	Adverse event	Adverse drug reaction	Adverse event	Adverse drug reaction
All events	21 (38.9)	1 (1.9)	24 (40.7)	2 (3.4)	26 (46.4)	1 (1.8)	24 (41.4)	0 (0.0)	28 (53.8)	3 (5.8)
Nasopharyngitis	13 (24.1)	0 (0.0)	12 (20.3)	0 (0.0)	10 (17.9)	0 (0.0)	7 (12.1)	0 (0.0)	12 (23.1)	0 (0.0)
Gastroenteritis	1 (1.9)	0 (0.0)	1 (1.7)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	3 (5.8)	0 (0.0)
Cystitis	1 (1.9)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.7)	0 (0.0)	2 (3.8)	1 (1.9)
Influenza	2 (3.7)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	2 (3.8)	0 (0.0)
Dental caries	1 (1.9)	0 (0.0)	2 (3.4)	0 (0.0)	0 (0.0)	0 (0.0)	2 (3.4)	0 (0.0)	1 (1.9)	0 (0.0)
Gingivitis	0 (0.0)	0 (0.0)	2 (3.4)	0 (0.0)	1 (1.8)	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.9)	0 (0.0)
Constipation	1 (1.9)	0 (0.0)	3 (5.1)	1 (1.7)	0 (0.0)	0 (0.0)	2 (3.4)	0 (0.0)	0 (0.0)	0 (0.0)
Reflux oesophagitis	2 (3.7)	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.8)	0 (0.0)	1 (1.7)	0 (0.0)	0 (0.0)	0 (0.0)
Cataract	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	2 (3.8)	0 (0.0)
Back pain	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	2 (3.6)	0 (0.0)	2 (3.4)	0 (0.0)	1 (1.9)	0 (0.0)
Headache	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	3 (5.4)	0 (0.0)	2 (3.4)	0 (0.0)	1 (1.9)	0 (0.0)
Upper respiratory tract inflammation	0 (0.0)	0 (0.0)	1 (1.7)	0 (0.0)	0 (0.0)	0 (0.0)	3 (5.2)	0 (0.0)	0 (0.0)	0 (0.0)
Eczema	3 (5.6)	1 (1.9)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)

Number of subjects with event (incidence %), MedDRA/J ver.13.0

One subject in the dapagliflozin 1 mg group died (cholecystitis/sepsis/multi-organ failure), but a causal relationship to the study drug was ruled out for the event. Four serious adverse events (cholecystitis/sepsis/multi-organ failure [fatal case], spinal compression fracture) occurred in 2 subjects in the dapagliflozin 1 mg group, 1 event (bladder cancer) in 1 subject in the dapagliflozin 2.5 mg group, 2 events (dermal cyst, acute myocardial infarction) in 2 subjects in the dapagliflozin 5 mg group, and 1 event (gastroenteritis) in 1 subject in the dapagliflozin 10 mg group, but a causal relationship to the study drug was ruled out for all these events. One adverse events leading to treatment discontinuation (cystitis bacterial) occurred in 1 subject in the dapagliflozin 1 mg group, 1 event (bladder cancer) in 1 subject in the dapagliflozin 2.5 mg group, and 1 event (acute myocardial infarction) in 1 subject in the dapagliflozin 5 mg group. The cystitis bacterial in the dapagliflozin 1 mg group was assessed as an adverse drug reaction.

Hypoglycaemia¹²⁹ was reported by 1 subject each in the placebo, dapagliflozin 2.5 mg, and 10 mg groups, but none of these events were severe.

Events suggestive of genital infection¹³⁰ were reported by 1 subject in the dapagliflozin 2.5 mg group (pruritus genital) and 1 subject in the dapagliflozin 5 mg group (balanitis).

Events suggestive of urinary tract infection¹³⁰ were reported by 1 subject in the placebo group (cystitis), 1 subject in the dapagliflozin 1 mg group (cystitis bacterial), 1 subject in the dapagliflozin 5 mg group (cystitis), and 2 subjects in the dapagliflozin 10 mg group (cystitis in 2 subjects).

Among haematological parameters, dose-dependent increases from baseline in mean haematocrit and haemoglobin values were observed in the dapagliflozin groups. Haematocrit abnormally high (>55%) was reported by 5 subjects in the dapagliflozin groups (1 subject in the 1 mg group, 1

¹²⁹ Definition of hypoglycaemia: (a) Severe hypoglycaemia, Any symptomatic event with blood glucose of <54 mg/dL that requires third-party assistance due to severe consciousness disturbed or behavior disorder and recovers immediately by treatment with glucose or glucagon; (b) Mild hypoglycaemia, Any symptomatic or asymptomatic event with blood glucose of <63 mg/dL that does not meet the criteria of severe hypoglycaemia; (c) Other hypoglycaemia, Any event reported as an event suggestive of hypoglycaemia without any documentation of blood glucose levels.

Events of hypoglycaemia were collected separately from adverse events.

¹³⁰ Collected based on the list of preferred terms predefined by the applicant.

subject in the 2.5 mg group, 2 subjects in the 5 mg group, 1 subject in the 10 mg group), and haemoglobin abnormally high (>18 g/dL) was reported by 8 subjects in the dapagliflozin groups (1 subject in the 1 mg group, 2 subjects in the 2.5 mg group, 2 subjects in the 5 mg group, 3 subjects in the 10 mg group) and 1 subject in the placebo group, but none of these were associated with thromboembolism.

No clinically relevant changes were seen in vital signs or ECG.

4.(iii).A.(3) Phase III studies

4.(iii).A.(3).1 Japanese phase III study (monotherapy) (5.3.5.1.2, Study D1692C00006 [February 2011 to March 2012])

A placebo-controlled, randomized, double-blind, parallel-group study was conducted to evaluate the efficacy and safety of dapagliflozin in Japanese patients with type 2 diabetes mellitus¹³¹ (target sample size of 255; 85 subjects per group).

This study consisted of the washout period (6 weeks), single-blind placebo run-in period (4 weeks), double-blind treatment period (24 weeks), and follow-up period (3 weeks).

Placebo or dapagliflozin at a dose of 5 or 10 mg was to be orally administered once daily for 24 weeks in the morning.

All of the 261 treated subjects (87 subjects in the placebo group, 86 subjects in the 5 mg group, 88 subjects in the 10 mg group) were included in the safety analysis set and FAS. FAS was used for the primary efficacy analysis. A total of 22 subjects discontinued the study, including 8 subjects in the placebo group (3 subjects due to an adverse event, 2 subjects each due to ineligibility and consent withdrawal, 1 subject due to poor compliance/non-compliance with the study protocol), 5 subjects in the dapagliflozin 5 mg group (2 subjects due to ineligibility, 1 subject each due to an adverse event, consent withdrawal, and poor compliance/non-compliance with the study protocol), and 9 subjects in the dapagliflozin 10 mg group (5 subjects due to ineligibility, 2 subjects each due to an adverse event and consent withdrawal). Initiation of hyperglycaemia rescue therapy¹³² was considered at the discretion of the investigator in subjects with 2 consecutive fasting blood glucose measurements of >200 mg/dL at Week 12 to 24 of treatment, but no subject actually received hyperglycaemia rescue therapy.

The primary efficacy endpoint of the change in HbA1c from baseline (start of the double-blind treatment period) to Week 24 of treatment in the FAS was as shown in Table 18. A statistically significant decrease was observed in the dapagliflozin 5 and 10 mg groups compared with the placebo group ($P < 0.0001$ for both comparisons, ANCOVA model at significance level of 2.7% [two-sided] for each comparison [Dunnett-adjusted significance level, overall significance level for the study was 5% (two-sided)]).

¹³¹ Key Inclusion Criteria were type 2 diabetes mellitus patients aged ≥ 20 years with a BMI of <45 kg/m² who met one of the following criteria: 1) Patients without a history of antidiabetic medication (with insulin or other hypoglycemic agents) at study entry and with HbA1c of $\geq 6.5\%$ and $\leq 10\%$ at study entry (washout not required); or 2) Patients with a previous or current history of treatment with drugs other than thiazolidines at study entry or within 6 weeks prior to study entry and with HbA1c of $\leq 8\%$ at study entry (washout required).

¹³² In principle, rescue therapy was to be performed with metformin, and if metformin use was not possible, glimepiride was to be used. Subjects who could receive neither of these drugs were to be withdrawn from the study. Dosage regimen was not specified.

Table 18. Change in HbA1c from baseline to Week 24 of treatment (Study D1692C00006, FAS)

	Placebo group (n = 87)	Dapagliflozin 5 mg group (n = 86)	Dapagliflozin 10 mg group (n = 88)
Baseline	7.50 ± 0.629	7.50 ± 0.718	7.46 ± 0.611
Week 24 ^{a)}	7.42 ± 0.848	7.08 ± 0.687	7.00 ± 0.538
Change from baseline ^{b)}	-0.06 [-0.18, 0.06]	-0.41 [-0.53, -0.29]	-0.45 [-0.57, -0.33]
Difference from the placebo group ^{b)}	-	-0.35 [-0.52, -0.18]	-0.39 [-0.56, -0.23]
P-value ^{b,c)}	-	<0.0001	<0.0001

Unit, %; mean ± SD; adjusted mean [two-sided 95% CI]; -, Not applicable

a) LOCF

b) ANCOVA model with treatment and sex as fixed effects and baseline HbA1c as a covariate

c) Multiplicity of statistical tests was adjusted by using Dunnett's method, and a significance level of 0.027 was used.

The time courses of the change in HbA1c from baseline to Week 24 of treatment were as shown in Figure 1.

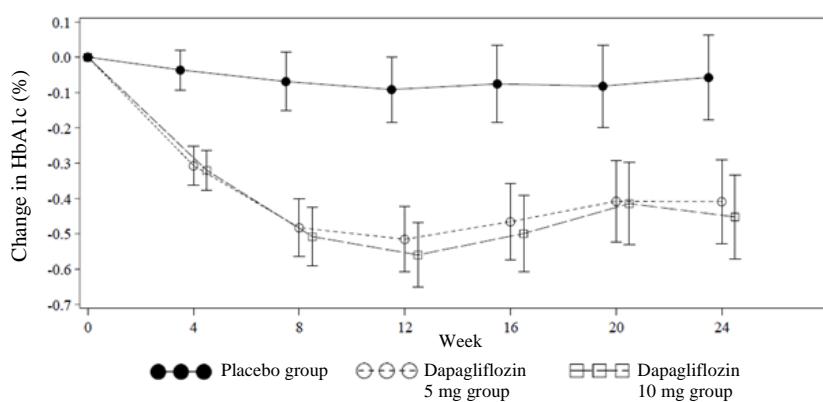


Figure 1. Time courses of the change in HbA1c from baseline to Week 24 of treatment (Adjusted mean and its two-sided 95% CI) (Study D1692C00006, FAS)

The results of analysis of key secondary endpoints were as shown in Table 19.

Table 19. Changes in fasting blood glucose and body weight from baseline to Week 24 of treatment (Study D1692C00006, FAS)

	Fasting blood glucose (mg/dL)			Body weight (kg)		
	Placebo group (n = 87)	Dapagliflozin 5 mg group (n = 86)	Dapagliflozin 10 mg group (n = 88)	Placebo group (n = 87)	Dapagliflozin 5 mg group (n = 86)	Dapagliflozin 10 mg group (n = 88)
Baseline	139.8 ± 21.71	137.5 ± 24.41	138.7 ± 22.26	65.96 ± 12.908	65.81 ± 14.371	69.70 ± 13.821
Week 24 ^{a)}	146.1 ± 26.67	130.2 ± 25.34	125.8 ± 19.39	65.31 ± 12.801	63.86 ± 14.706	67.54 ± 13.249
Change from baseline ^{b)}	5.8 [1.6, 10.1]	-8.6 [-12.9, -4.3]	-13.7 [-18.0, -9.5]	-0.84 [-1.36, -0.32]	-2.13 [-2.65, -1.60]	-2.22 [-2.73, -1.71]
Difference from the placebo group ^{b)}	-	-14.4 [-20.1, -8.7]	-19.5 [-25.2, -13.8]	-	-1.29 [-1.98, -0.59]	-1.38 [-2.08, -0.69]

Mean ± SD, adjusted mean [two-sided 95% CI]; -, Not applicable

a) LOCF

b) ANCOVA model with treatment and sex as fixed effects and baseline values as covariates

Regarding safety, the incidences of adverse events¹³³ and adverse drug reactions were respectively 51.7% (45 of 87 subjects) and 13.8% (12 of 87 subjects) in the placebo group, 47.7%

¹³³ Events that occurred during the treatment period or within 4 days after the end of treatment.

(41 of 86 subjects) and 7.0% (6 of 86 subjects) in the dapagliflozin 5 mg group, and 64.8% (57 of 88 subjects) and 19.3% (17 of 88 subjects) in the dapagliflozin 10 mg group. Adverse events and adverse drug reactions reported by $\geq 3\%$ of subjects in any treatment group were as shown in Table 20.

Table 20. Adverse events and adverse drug reactions reported by $\geq 3\%$ of subjects in any treatment group (Study D1692C00006, safety analysis set)

Event term	Placebo group (n = 87)		Dapagliflozin 5 mg group (n = 86)		Dapagliflozin 10 mg group (n = 88)	
	Adverse event	Adverse drug reaction	Adverse event	Adverse drug reaction	Adverse event	Adverse drug reaction
All events	45 (51.7)	12 (13.8)	41 (47.7)	6 (7.0)	57 (64.8)	17 (19.3)
Nasopharyngitis	9 (10.3)	0 (0.0)	9 (10.5)	0 (0.0)	15 (17.0)	0 (0.0)
Dental caries	1 (1.1)	0 (0.0)	0 (0.0)	0 (0.0)	4 (4.5)	0 (0.0)
Pollakiuria	1 (1.1)	1 (1.1)	2 (2.3)	2 (2.3)	4 (4.5)	4 (4.5)
Renal impairment	3 (3.4)	2 (2.3)	2 (2.3)	0 (0.0)	4 (4.5)	1 (1.1)
Back pain	1 (1.1)	0 (0.0)	0 (0.0)	0 (0.0)	3 (3.4)	1 (1.1)
Hypertension	5 (5.7)	1 (1.1)	2 (2.3)	1 (1.2)	1 (1.1)	1 (1.1)

Number of subjects with event (incidence %), MedDRA/J ver.14.1

No deaths were reported. Serious adverse events were reported by 1 subject in the placebo group (putamen haemorrhage) and 1 subject in the dapagliflozin 10 mg group (rib fracture), but a causal relationship to the study drug was ruled out for both events. Adverse events leading to treatment discontinuation were reported by 5 subjects in the placebo group (diabetes mellitus and renal impairment in 2 subjects each, putamen haemorrhage in 1 subject); 3 subjects in the dapagliflozin 5 mg group (renal impairment in 2 subjects, cholelithiasis in 1 subject); and 7 subjects in the dapagliflozin 10 mg group (renal impairment in 3 subjects, glomerular filtration rate decreased in 2 subjects, hypertension and urinary tract infection in 1 subject each). Among these, diabetes mellitus (2 subjects) and renal impairment (1 subject) in the placebo group, cholelithiasis (1 subject) in the dapagliflozin 5 mg group, and hypertension, renal impairment, and urinary tract infection (1 subject each) in the dapagliflozin 10 mg group were assessed as adverse drug reactions.

Hypoglycaemia¹²⁹ was reported by 2 subjects in the dapagliflozin 10 mg group, but no severe hypoglycaemia was reported.

Events suggestive of genital infection¹³⁰ were reported by 1 subject in the placebo group (pruritus genital), 1 subject in the dapagliflozin 5 mg group (vulvovaginal candidiasis), and 2 subjects in the dapagliflozin 10 mg group (pruritus genital, vulvitis).

Events suggestive of urinary tract infection¹³⁰ were reported by 2 subjects in the placebo group (cystitis) and 2 subjects in the dapagliflozin 10 mg group (cystitis, urinary tract infection).

Among haematological parameters, haematocrit abnormally high and haemoglobin abnormally high were reported by 1 subject in the dapagliflozin 5 mg group, but these were not associated with thromboembolism.

Among vital signs, the dapagliflozin groups showed a slightly-greater decrease in sitting systolic blood pressure from baseline at Week 24 of treatment as compared with the placebo group (-1.966 mmHg in the placebo group, -2.705 mmHg in the dapagliflozin 5 mg group, -3.439 mmHg in the dapagliflozin 10 mg group), but this parameter returned to baseline during the follow-up period. No clinically meaningful changes were observed in sitting diastolic blood pressure. No changes were observed in ECG parameters from baseline through Week 24 of treatment in most of the subjects.

4.(iii).A.(3).2 Japanese phase III long-term treatment study (monotherapy and concomitant therapy) (5.3.5.2.1, Study D1692C00012 [February 2011 to September 2012])

An open-label, uncontrolled, long-term treatment study was conducted to evaluate the safety and efficacy of monotherapy and concomitant therapies with dapagliflozin in Japanese patients with type 2 diabetes mellitus¹³⁴ which has not adequately responded to diet and/or exercise therapy or any of the following hypoglycemic agents: sulfonylureas (SUs), fast-acting insulin secretagogues (glinides), biguanides (BGs), α -glucosidase inhibitors (α -GIs), thiazolidines (TZDs), dipeptidyl peptidase-4 inhibitors (DPP-4), or glucagon-like peptide-1 receptor agonists (GLP-1) (target sample size of 700; 240 subjects in the monotherapy group, 460 subjects in the concomitant therapy groups [120 subjects in the dapagliflozin + SU group, 60 subjects each in the dapagliflozin + DPP-4, α -GI, BG, and TZD groups, 50 subjects each in the dapagliflozin + glinide and GLP-1 groups]).

This study consisted of the washout period (6 weeks), run-in period (4 weeks), treatment period (52 weeks), and follow-up period (3 weeks).

Dapagliflozin 5 mg was to be orally administered once daily in the morning. From Week 12 of treatment, the dose was allowed to be increased to 10 mg from the subsequent visit (during or after Week 16) in subjects with HbA1c of >7.5% and no safety concerns. In principle, subjects for whom the dose was increased to 10 mg were not allowed to have dose reduction to 5 mg thereafter, and those requiring dose reduction as determined by the investigator were to be withdrawn from the study. In the concomitant therapy groups, the dosage regimens of hypoglycemic agents were the same as those used before study treatment; excluding, however, SUs for which dose reduction was permitted.¹³⁵ In subjects with HbA1c of >8% at clinic visits between Weeks 24 and 52 of treatment in spite of 8-week treatment with dapagliflozin at the increased dose of 10 mg, initiation of hyperglycaemia rescue therapy¹³⁶ was considered at the subsequent visits at the discretion of the investigator.

All of the following 728 treated subjects were included in the safety analysis set: 249 subjects in the monotherapy group, 479 subjects in the concomitant therapy groups (122 subjects in the dapagliflozin + SU group,¹³⁷ 62 subjects in the dapagliflozin + DPP-4 group,¹³⁸ 61 subjects in

¹³⁴ Key Inclusion Criteria were type 2 diabetes mellitus patients aged \geq 20 years with BMI $<$ 45 kg/m² who met one of the following criteria: 1) Monotherapy group, Patients without a history of antidiabetic medication at study entry and with HbA1c of \geq 6.5% and \leq 10% at study entry (washout not required), or patients with a previous or current history of treatment with drugs other than TZDs during or within 6 weeks prior to study entry and with HbA1c of \leq 8% at study entry (washout required); 2) Dapagliflozin + SU, glinide, BG, α -GI, DPP-4, or GLP-1 group, Patients with a history of treatment with hypoglycemic agent(s) (as basal agent(s)) each at a constant dose within the approved dose range for \geq 8 weeks prior to the start of dapagliflozin and with HbA1c of \geq 6.5% and \leq 10% at study entry; or 3) Dapagliflozin + TZD group, Patients with a history of treatment with pioglitazone hydrochloride at a constant dose within the approved dose range for \geq 12 weeks prior to the start of dapagliflozin and with HbA1c of \geq 6.5% and \leq 10% at study entry.

¹³⁵ The criteria for dose reduction of SUs were defined as follows: The investigator should reduce the dose of SUs for subjects with at least 2 measurements of blood glucose of \leq 70 mg/dL at any visit after the start of treatment. No subjects in the dapagliflozin + SU group had dose reduction of SU during the period of 52 weeks.

¹³⁶ Subjects in the monotherapy group were allowed to receive another hypoglycemic agent at a gradually increasing dose up to the maximum approved dose. In principle, subjects in the concomitant therapy groups were required to receive a gradual dose increase of the basal agent(s) up to the maximum approved dose, and as needed, allowed to receive another hypoglycemic agent at a gradually increasing dose up to the maximum approved dose.

¹³⁷ As a concomitant drug, glimepiride was used at the daily dose of 0.5 mg in 2 subjects, 1 mg in 67 subjects, 1.5 mg in 3 subjects, 2 mg in 23 subjects, 2.5 mg in 1 subject, 3 mg in 14 subjects, 4 mg in 5 subjects, 5 mg in 1 subject, and 6 mg in 6 subjects.

¹³⁸ As a concomitant drug, sitagliptin phosphate hydrate (sitagliptin) was used at the daily dose of 25 mg in 2 subjects, 50 mg in 46 subjects, and 100 mg in 14 subjects.

the dapagliflozin + α -GI group,¹³⁹ 71 subjects in the dapagliflozin + BG group,¹⁴⁰ 64 subjects in the dapagliflozin + TZD group,¹⁴¹ 49 subjects in the dapagliflozin + glinide group,¹⁴² 50 subjects in the dapagliflozin + GLP-1 group¹⁴³). Of these, the following 726 subjects¹⁴⁴ were included in the FAS: 249 subjects in the monotherapy group, 477 subjects in the concomitant therapy groups (122 subjects in the dapagliflozin + SU group, 62 subjects in the dapagliflozin + DPP-4 group, 61 subjects in the dapagliflozin + α -GI group, 69 subjects in the dapagliflozin + BG group, 64 subjects in the dapagliflozin + TZD group, 49 subjects in the dapagliflozin + glinide group, 50 subjects in the dapagliflozin + GLP-1 group), and the FAS was used for the efficacy analysis. A total of 630 subjects (86.5%) completed the 52-week treatment period. A total of 98 subjects discontinued the study, including 28 subjects in the monotherapy group¹⁴⁵ and 70 subjects in the concomitant therapy groups.¹⁴⁶ A total of 52 subjects¹⁴⁷ received hyperglycaemia rescue therapy during 52 weeks of treatment.

No primary efficacy endpoints were specified. The secondary endpoint of the change in HbA1c from baseline to Week 52 of treatment in the FAS was as shown in Table 21, and the time courses of the change in HbA1c from baseline to Week 52 of treatment were as shown in Figure 2.

Table 21. Change in HbA1c from baseline to Week 52 of treatment (excluding data from subjects after hyperglycaemia rescue therapy) (Study D1692C00012, FAS)

	Monotherapy group (n = 249)	Dapagliflozin + SU group (n = 122)	Dapagliflozin + DPP-4 group (n = 62)	Dapagliflozin + α -GI group (n = 61)	Dapagliflozin + BG group (n = 69)	Dapagliflozin + TZD group (n = 64)	Dapagliflozin + glinide group (n = 49)	Dapagliflozin + GLP-1 group (n = 50)
Baseline	7.53 ± 0.761	8.02 ± 0.842	7.80 ± 0.909	7.59 ± 0.729	7.63 ± 0.845	7.94 ± 0.915	7.49 ± 0.725	8.11 ± 0.917
Week 52 ^{a)}	6.87 ± 0.717	7.37 ± 0.823	7.20 ± 0.826	6.78 ± 0.637	6.99 ± 0.727	7.08 ± 0.693	6.73 ± 0.513	7.62 ± 1.009
Change from baseline	-0.66 [-0.75, -0.57]	-0.65 [-0.78, -0.52]	-0.60 [-0.74, -0.45]	-0.81 [-0.98, -0.63]	-0.63 [-0.80, -0.47]	-0.86 [-1.05, -0.67]	-0.76 [-0.95, -0.57]	-0.49 [-0.72, -0.26]

Unit, %; mean ± SD; mean [two-sided 95% CI]

a) LOCF

¹³⁹ As a concomitant drug, voglibose was used in 32 subjects (at the daily dose of 0.6 mg in 16 subjects and 0.9 mg in 16 subjects), miglitol in 26 subjects (at the daily dose of 150 mg in 23 subjects and 225 mg in 3 subjects), and acarbose in 3 subjects (at the daily dose of 150 mg in 1 subject and 300 mg in 2 subjects).

¹⁴⁰ As a concomitant drug, metformin hydrochloride (metformin) was used at the daily dose of 500 mg in 18 subjects, 750 mg in 38 subjects, 1000 mg in 3 subjects, and 1500 mg in 12 subjects.

¹⁴¹ As a concomitant drug, pioglitazone hydrochloride (pioglitazone) was used at the daily dose of 15 mg in 32 subjects, 30 mg in 31 subjects, and 45 mg in 1 subject.

¹⁴² As a concomitant drug, nateglinide was used at the daily dose of 270 mg in 27 subjects, and mitiglinide calcium hydrate (mitiglinide) at the daily dose of 30 mg in 22 subjects.

¹⁴³ As a concomitant drug, liraglutide (genetical recombination) (liraglutide) was used at the daily dose of 0.6 mg in 7 subjects and 0.9 mg in 43 subjects.

¹⁴⁴ Consisting of subjects who had baseline value and at least 1 post-dose value for at least 1 efficacy variable.

¹⁴⁵ Including 11 subjects due to consent withdrawal, 10 subjects due to adverse events, 5 subjects due to ineligibility, and 2 subjects due to poor compliance/non-compliance with the study protocol.

¹⁴⁶ Including 18 subjects in the dapagliflozin + SU group (7 subjects due to adverse events, 5 subjects each due to ineligibility and consent withdrawal, 1 subject due to poor compliance/non-compliance with the study protocol), 9 subjects in the dapagliflozin + DPP-4 group (5 subjects due to consent withdrawal, 3 subjects due to adverse events, 1 subject due to poor compliance/non-compliance with the study protocol), 8 subjects in the dapagliflozin + α -GI group (4 subjects due to consent withdrawal, 2 subjects due to adverse events, 1 subject each due to ineligibility and poor compliance/non-compliance with the study protocol), 15 subjects in the dapagliflozin + BG group (9 subjects due to ineligibility, 4 subjects due to consent withdrawal, 2 subjects due to adverse events), 9 subjects in the dapagliflozin + TZD group (5 subjects due to consent withdrawal, 3 subjects due to ineligibility, 1 subject due to poor compliance/non-compliance with the study protocol), 4 subjects in the dapagliflozin + glinide group (2 subjects due to adverse events, 1 subject each due to consent withdrawal and poor compliance/non-compliance with the study protocol), and 7 subjects in the dapagliflozin + GLP-1 group (4 subjects due to consent withdrawal, 2 subjects due to ineligibility, 1 subject due to adverse events).

¹⁴⁷ Including 4 subjects in the monotherapy group, 17 subjects in the dapagliflozin + SU group, 7 subjects in the dapagliflozin + DPP-4 group, 5 subjects in the dapagliflozin + BG group, 2 subjects in the dapagliflozin + TZD group, 1 subject in the dapagliflozin + glinide group, and 16 subjects in the dapagliflozin + GLP-1 group. One subject in the dapagliflozin + SU group discontinued the study due to hyperglycaemia before receiving rescue therapy.

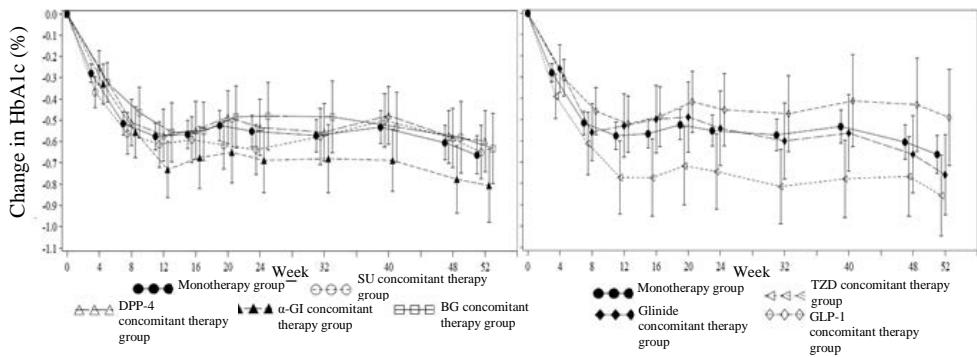


Figure 2. Time courses of the change in HbA1c from baseline to Week 52 of treatment (LOCF; excluding data from subjects after hyperglycaemia rescue therapy) (Adjusted mean and its two-sided 95% CI) (Study D1692C00012, FAS)

The results of analysis of key secondary endpoints were as shown in Table 22.

Table 22. Results of analysis of key secondary endpoints (excluding data from subjects after hyperglycaemia rescue therapy) (Study D1692C00012, FAS)

		Monotherapy group (n = 249)	Dapagliflozin + SU group (n = 122)	Dapagliflozin + DPP-4 group (n = 62)	Dapagliflozin + α-GI group (n = 61)	Dapagliflozin + BG group (n = 69)	Dapagliflozin + TZD group (n = 64)	Dapagliflozin + glinide group (n = 49)	Dapagliflozin + GLP-1 group (n = 50)
Fasting blood glucose (mg/dL)	No. of subjects	n = 245	n = 120	n = 59	n = 61	n = 66	n = 64	n = 49	n = 50
	Baseline	140.1 ± 24.8	149.9 ± 29.7	147.5 ± 23.7	141.6 ± 25.0	148.4 ± 32.5	144.1 ± 30.8	147.8 ± 30.1	150.2 ± 29.2
	Week 52 ^{a)}	125.7 ± 18.7	131.5 ± 24.2	129.8 ± 17.6	124.9 ± 18.5	130.6 ± 24.3	126.6 ± 19.9	126.9 ± 19.7	138.7 ± 30.2
	Change [two-sided 95% CI]	-14.3 [-17.0, -11.6]	-18.4 [-23.1, -13.7]	-17.6 [-23.9, -11.4]	-16.6 [-22.6, -10.7]	-17.8 [-25.2, -10.3]	-17.5 [-23.8, -11.2]	-20.9 [-27.8, -13.9]	-11.5 [-20.1, -2.9]
Body weight (kg)	No. of subjects	n = 249	n = 122	n = 62	n = 61	n = 69	n = 64	n = 49	n = 50
	Baseline	67.8 ± 13.4	66.1 ± 12.1	64.7 ± 11.8	69.3 ± 16.7	68.4 ± 14.2	72.7 ± 16.7	67.2 ± 13.8	63.4 ± 16.7
	Week 52 ^{a)}	65.2 ± 13.0	64.4 ± 12.1	62.3 ± 11.3	66.9 ± 17.3	66.2 ± 14.0	72.0 ± 17.3	64.7 ± 13.8	60.5 ± 14.6
	Change [two-sided 95% CI]	-2.58 [-2.87, -2.30]	-1.75 [-2.19, -1.32]	-2.42 [-2.86, -1.97]	-2.44 [-3.22, -1.65]	-2.25 [-2.73, -1.76]	-0.77 [-1.49, -0.04]	-2.47 [-3.09, -1.86]	-2.90 [-4.14, -1.65]

Mean ± SD, mean [two-sided 95% CI]

a) LOCF

Regarding safety, the incidences of adverse events and adverse drug reactions were, in this order, as follows: 79.1% (197 of 249 subjects) and 24.9% (62 of 249 subjects) in the monotherapy group; 73.0% (89 of 122 subjects) and 16.4% (20 of 122 subjects) in the dapagliflozin + SU group; 75.8% (47 of 62 subjects) and 17.7% (11 of 62 subjects) in the dapagliflozin + DPP-4 group; 63.9% (39 of 61 subjects) and 11.5% (7 of 61 subjects) in the dapagliflozin + α-GI group; 78.9% (56 of 71 subjects) and 28.2% (20 of 71 subjects) in the dapagliflozin + BG group; 70.3% (45 of 64 subjects) and 12.5% (8 of 64 subjects) in the dapagliflozin + TZD group; 69.4% (34 of 49 subjects) and 20.4% (10 of 49 subjects) in the dapagliflozin + glinide group; and 74.0% (37 of 50 subjects) and 16.0% (8 of 50 subjects) in the dapagliflozin + GLP-1 group. Adverse events reported by ≥5% of subjects in any treatment group were as shown in Table 23, and adverse drug reactions reported by ≥3% of subjects in any treatment group were as shown in Table 24.

**Table 23. Adverse events reported by ≥5% of subjects in any treatment group
(Study D1692C00012, safety analysis set)**

Event term	Monotherapy group (n = 249)	Dapagliflozin + SU group (n = 122)	Dapagliflozin + DPP-4 group (n = 62)	Dapagliflozin + α-GI group (n = 61)	Dapagliflozin + BG group (n = 71)	Dapagliflozin + TZD group (n = 64)	Dapagliflozin + glinide group (n = 49)	Dapagliflozin + GLP-1 group (n = 50)
All events	197 (79.1)	89 (73.0)	47 (75.8)	39 (63.9)	56 (78.9)	45 (70.3)	34 (69.4)	37 (74.0)
Nasopharyngitis	63 (25.3)	33 (27.0)	10 (16.1)	12 (19.7)	14 (19.7)	18 (28.1)	19 (38.8)	10 (20.0)
Upper respiratory tract infection	12 (4.8)	1 (0.8)	6 (9.7)	0 (0.0)	1 (1.4)	2 (3.1)	0 (0.0)	0 (0.0)
Pharyngitis	9 (3.6)	3 (2.5)	3 (4.8)	4 (6.6)	2 (2.8)	3 (4.7)	0 (0.0)	0 (0.0)
Gastroenteritis	6 (2.4)	3 (2.5)	5 (8.1)	0 (0.0)	1 (1.4)	3 (4.7)	1 (2.0)	1 (2.0)
Bronchitis	5 (2.0)	3 (2.5)	5 (8.1)	1 (1.6)	2 (2.8)	4 (6.3)	0 (0.0)	1 (2.0)
Constipation	9 (3.6)	3 (2.5)	3 (4.8)	1 (1.6)	7 (9.9)	1 (1.6)	4 (8.2)	3 (6.0)
Gastritis	4 (1.6)	1 (0.8)	5 (8.1)	1 (1.6)	0 (0.0)	1 (1.6)	0 (0.0)	1 (2.0)
Diarrhoea	2 (0.8)	1 (0.8)	1 (1.6)	4 (6.6)	2 (2.8)	1 (1.6)	1 (2.0)	0 (0.0)
Gastrooesophageal reflux disease	1 (0.4)	2 (1.6)	0 (0.0)	0 (0.0)	1 (1.4)	0 (0.0)	0 (0.0)	5 (10.0)
Back pain	10 (4.0)	9 (7.4)	4 (6.5)	1 (1.6)	3 (4.2)	2 (3.1)	2 (4.1)	0 (0.0)
Arthralgia	7 (2.8)	6 (4.9)	0 (0.0)	0 (0.0)	4 (5.6)	2 (3.1)	0 (0.0)	0 (0.0)
Pollakiuria	13 (5.2)	2 (1.6)	1 (1.6)	2 (3.3)	3 (4.2)	3 (4.7)	2 (4.1)	0 (0.0)
Renal impairment	4 (1.6)	1 (0.8)	0 (0.0)	0 (0.0)	7 (9.9)	1 (1.6)	1 (2.0)	2 (4.0)
Insomnia	2 (0.8)	2 (1.6)	1 (1.6)	1 (1.6)	0 (0.0)	1 (1.6)	2 (4.1)	4 (8.0)

Number of subjects with event (incidence %), MedDRA/J ver.15.0

**Table 24. Adverse drug reactions reported by ≥3% of subjects in any treatment group
(Study D1692C00012, safety analysis set)**

Event term	Monotherapy group (n = 249)	Dapagliflozin + SU group (n = 122)	Dapagliflozin + DPP-4 group (n = 62)	Dapagliflozin + α-GI group (n = 61)	Dapagliflozin + BG group (n = 71)	Dapagliflozin + TZD group (n = 64)	Dapagliflozin + glinide group (n = 49)	Dapagliflozin + GLP-1 group (n = 50)
All events	62 (24.9)	20 (16.4)	11 (17.7)	7 (11.5)	20 (28.2)	8 (12.5)	10 (20.4)	8 (16.0)
Pollakiuria	13 (5.2)	2 (1.6)	1 (1.6)	1 (1.6)	3 (4.2)	3 (4.7)	2 (4.1)	0 (0.0)
Renal impairment	1 (0.4)	0 (0.0)	0 (0.0)	0 (0.0)	3 (4.2)	0 (0.0)	1 (2.0)	0 (0.0)
Thirst	9 (3.6)	2 (1.6)	0 (0.0)	2 (3.3)	1 (1.4)	1 (1.6)	1 (2.0)	0 (0.0)
Malaise	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.6)	0 (0.0)	2 (4.0)
Constipation	3 (1.2)	1 (0.8)	2 (3.2)	1 (1.6)	4 (5.6)	1 (1.6)	1 (2.0)	0 (0.0)
Vulvovaginal pruritus	1 (0.4)	0 (0.0)	2 (3.2)	0 (0.0)	0 (0.0)	1 (1.6)	1 (2.0)	0 (0.0)

Number of subjects with event (incidence %), MedDRA/J ver.15.0

No deaths were reported. Serious adverse events were reported by 14 subjects in the monotherapy group (colon cancer and colonic polyp in 2 subjects each, age-related macular degeneration, cataract, retinal detachment, breast cancer, chronic sinusitis, diverticulitis, brain stem haemorrhage, dizziness, limb traumatic amputation, and osteoarthritis in 1 subject each); 5 subjects in the dapagliflozin + SU group (cerebral infarction in 2 subjects, rectal cancer, lobar pneumonia, and spinal compression fracture in 1 subject each); 3 subjects in the dapagliflozin + DPP-4 group (cataract, haemorrhoids, tachycardia paroxysmal); 2 subjects in the dapagliflozin + α-GI group (pharyngitis, electrocardiogram abnormal); 2 subjects in the dapagliflozin + BG group (cholelithiasis, calculus urinary); 1 subject in the dapagliflozin + TZD group (vertigo positional); 1 subject in the dapagliflozin + glinide group (gastroenteritis); and 1 subject in the dapagliflozin + GLP-1 group (cerebral infarction/gastrointestinal inflammation). Colon cancer and breast cancer in the monotherapy group, cerebral infarction in the dapagliflozin + SU group, calculus urinary in the dapagliflozin + BG group, and vertigo positional in the dapagliflozin + TZD group were assessed as adverse drug reactions. Adverse events leading to treatment discontinuation were reported by 15 subjects in the monotherapy group (renal impairment in 4 subjects, colon cancer in 2 subjects, thirst, glomerular filtration rate decreased, dizziness, breast cancer, dyspepsia, blood pressure increased, muscular weakness, hepatic function abnormal, and diabetes mellitus in 1 subject each), 8 subjects in the dapagliflozin + SU group (nasopharyngitis, renal impairment, lobar pneumonia, cerebral infarction, alcoholic liver disease, rectal cancer, intracranial aneurysm,

rash pruritic); 3 subjects in the dapagliflozin + DPP-4 group (eczema, vulvovaginal pruritus, extraocular muscle paresis); 1 subject in the dapagliflozin + α -GI group (dysuria); 11 subjects in the dapagliflozin + BG group (renal impairment in 6 subjects, glomerular filtration rate decreased in 3 subjects, hypertriglyceridaemia and cholelithiasis in 1 subject each); 2 subjects in the dapagliflozin + TZD group (renal impairment, renal failure); 2 subjects in the dapagliflozin + glinide group (liver function test abnormal, palpitations); and 3 subjects in the dapagliflozin + GLP-1 group (renal impairment in 2 subjects, drug eruption in 1 subject). Thirst, breast cancer, dyspepsia, muscular weakness, renal impairment (1 subject), and rectal cancer in the monotherapy group, cerebral infarction, alcoholic liver disease, intracranial aneurysm, and rash pruritic in the dapagliflozin + SU group, eczema, vulvovaginal pruritus, and extraocular muscle paresis in the dapagliflozin + DPP-4 group, dysuria in the dapagliflozin + α -GI group, renal impairment (2 subjects) in the dapagliflozin + BG group, and drug eruption in the dapagliflozin + GLP-1 group were assessed as adverse drug reactions.

The incidence of hypoglycaemia¹²⁹ was 2.4% (6 of 249 subjects) in the monotherapy group; 6.6% (8 of 122 subjects) in the dapagliflozin + SU group; 3.2% (2 of 62 subjects) in the dapagliflozin + DPP-4 group; 0.0% (0 of 61 subjects) in the dapagliflozin + α -GI group; 2.8% (2 of 71 subjects) in the dapagliflozin + BG group; 1.6% (1 of 64 subjects) in the dapagliflozin + TZD group; 6.1% (3 of 49 subjects) in the dapagliflozin + glinide group; and 6.0% (3 of 50 subjects) in the dapagliflozin + GLP-1 group. No severe events of hypoglycaemia were reported.

The incidence of events suggestive of genital infection¹³⁰ was 4.0% (10 of 249 subjects) in the monotherapy group; 3.3% (4 of 122 subjects) in the dapagliflozin + SU group; 6.5% (4 of 62 subjects) in the dapagliflozin + DPP-4 group; 1.6% (1 of 61 subjects) in the dapagliflozin + α -GI group; 4.2% (3 of 71 subjects) in the dapagliflozin + BG group; 1.6% (1 of 64 subjects) in the dapagliflozin + TZD group; 6.1% (3 of 49 subjects) in the dapagliflozin + glinide group; and 4.0% (2 of 50 subjects) in the dapagliflozin + GLP-1 group.

The incidence of events suggestive of urinary tract infection¹³⁰ was 3.6% (9 of 249 subjects) in the monotherapy group; 3.3% (4 of 122 subjects) in the dapagliflozin + SU group; 1.6% (1 of 62 subjects) in the dapagliflozin + DPP-4 group; 3.3% (2 of 61 subjects) in the dapagliflozin + α -GI group; 2.8% (2 of 71 subjects) in the dapagliflozin + BG group; 4.7% (3 of 64 subjects) in the dapagliflozin + TZD group; 0.0% (0 of 49 subjects) in the dapagliflozin + glinide group; and 4.0% (2 of 50 subjects) in the dapagliflozin + GLP-1 group.

Among hematological parameters, at Week 52 of treatment, haematocrit high (>55%) was reported by 8 subjects (3 subjects in the monotherapy group, 2 subjects in the dapagliflozin + SU group, 1 subject in the dapagliflozin + α -GI group, 2 subjects in the dapagliflozin + GLP-1 group), and haemoglobin high (>18 g/dL) was reported by 9 subjects (4 subjects in the monotherapy group, 3 subjects in the dapagliflozin + SU group, 1 subject in the dapagliflozin + α -GI group, 1 subject in the dapagliflozin + GLP-1 group), but thromboembolism-related adverse events (e.g., transient ischaemic attack, stroke, venous thromboembolism) associated with these findings were not reported. Among clinical chemistry parameters, alanine aminotransferase (ALT) abnormally high or aspartate aminotransferase (AST) abnormally high (>3-fold the upper limit of normal) was reported by 6 subjects (1 subject each in the monotherapy, and dapagliflozin + SU, DPP-4, α -GI, glinide, and GLP-1 groups), but no subjects reported ALT abnormally high or AST abnormally high that exceeded 5-fold the upper limit of normal. ALP abnormally high (>1.5-fold the upper limit of normal) was reported by 3 subjects (2 subjects in the monotherapy group, 1 subject in the dapagliflozin + GLP-1 group). Serum creatinine abnormally high (>1.5-fold the baseline) was reported by 1 subject in the dapagliflozin + SU group.

Among vital signs, a slight decrease from baseline in the mean sitting systolic blood pressure was observed 52 weeks after the start of treatment in all groups except the dapagliflozin + GLP-1

group. No clinically meaningful changes were observed in mean sitting diastolic blood pressure or mean sitting heart rate. ECG parameters were normal at baseline in 83.8% of subjects and remained normal after 24 and 52 weeks after the start of treatment in many of them.

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

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

The applicant explained as follows:

Dapagliflozin is a selective SGLT2 inhibitor with urinary glucose excretion promoting activity, a novel mechanism of action independent from insulin action, and therefore, is expected to have a low risk of hypoglycaemia and to reduce body weight by enhancing glucose excretion. In addition, with its efficacy and safety demonstrated not only as monotherapy but also as concomitant therapies with other antidiabetic drugs, dapagliflozin is expected to become a new therapeutic option for diabetes mellitus.

Because the clinical studies have confirmed the efficacy and safety of dapagliflozin as monotherapy and concomitant therapies [see “4.(iii).B.(2) Efficacy” and “4.(iii).B.(3) Safety”], PMDA considers that dapagliflozin can be a new therapeutic option for type 2 diabetes mellitus.

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

4.(iii).B.(2).1 Efficacy of monotherapy

PMDA asked the applicant to explain the reason why the difference from placebo in the change from baseline in HbA1c observed in a Japanese phase III study (Study D1692C00006) was smaller than that observed in the Japanese phase II study (Study D1692C00005).

The applicant responded as follows:

In Study D1692C00006, the difference [two-sided 95% CI] (%) in the change from baseline in HbA1c between the dapagliflozin and placebo groups was -0.42 [-0.55, -0.29] in the 5 mg group and -0.47 [-0.60, -0.34] in the 10 mg group at Week 12, and -0.35 [-0.52, -0.18] in the 5 mg group and -0.39 [-0.56, -0.23] in the 10 mg group at Week 24. The corresponding difference (%) observed in Study D1692C00005 was -0.74 [-0.93, -0.54] in the 5 mg group and -0.80 [-1.00, -0.61] in the 10 mg group at Week 12, showing smaller differences also in Study D1692C00006. An investigation of its reason revealed that Study D1692C00005 enrolled subjects with HbA1c of $\geq 7\%$ and $\leq 10\%$ and with eGFR¹⁴⁸ (mL/min/1.73 m²) of ≥ 60 at study entry, while Study D1692C00006 enrolled subjects with HbA1c of $\geq 6.5\%$ and $\leq 10\%$ and with eGFR of ≥ 45 at study entry, and thus, in the latter study, subjects with HbA1c of $<7\%$ and subjects with eGFR of <60 accounted for approximately 25% and 27.6% of the study population, respectively.

In Study D1692C00006, the differences [two-sided 95% CI] (%) in the change in HbA1c from baseline to Week 24 of treatment between the dapagliflozin 5 mg and placebo groups and between the dapagliflozin 10 mg and placebo groups were -0.14 [-0.48, 0.20] (n = 20) and -0.11 [-0.45, 0.24] (n = 19), respectively, in subjects with baseline HbA1c¹⁴⁹ of $<7\%$; -0.21 [-0.44, 0.01] (n = 50) and -0.26 [-0.49, -0.04] (n = 49), respectively, in subjects with baseline HbA1c of $\geq 7\%$ and $<8\%$; and -1.02 [-1.39, -0.65] (n = 16) and -0.95 [-1.30, -0.60] (n = 19), respectively, in subjects with baseline HbA1c of $\geq 8\%$; showing a greater change in HbA1c in subjects with higher baseline HbA1c. Similarly, the corresponding differences (%) were -0.37 [-0.68, -0.05] (n = 23) and -0.21 [-0.53, 0.10] (n = 24), respectively, in subjects with baseline eGFR of ≥ 45 and <60 ; and -0.37 [-0.57, -0.16] (n = 61) and -0.49 [-0.70, -0.29] (n = 61), respectively, in subjects with baseline eGFR of ≥ 60 and <90 ; showing a smaller change in HbA1c in subjects with lower baseline eGFR.

¹⁴⁸ Glomerular filtration rate estimating equations for Japanese patients (eGFR [mL/min/1.73 m²] = 194 × serum creatinine^{-1.094} × age^{-0.287} [for females, 194 × serum creatinine^{-1.094} × age^{-0.287} × 0.739]) was used in Studies D1692C00006 and D1692C00012, and the MDRD equation was used in Japanese Study D1692C00005 for calculation. The MDRD equation was used for calculation in foreign studies and updated pooled data analyses.

¹⁴⁹ Subjects with measurements of HbA1c both at baseline and at Week 24 are included.

As described above, the smaller overall change in HbA1c in Study D1692C00006 can be explained by the greater number of subjects with low baseline HbA1c and eGFR included in this study compared with Study D1692C00005.

However, given the facts that Study D1692C00006 confirmed the superiority of dapagliflozin at doses of 5 and 10 mg over placebo in the change in HbA1c from baseline to Week 24 of treatment (the primary endpoint) and that the Japanese phase III long-term treatment study (Study D1692C00012) demonstrated persistence of efficacy for up to Week 52 as evidenced by the change in HbA1c from baseline to Week 52 of treatment in the monotherapy group of -0.66 [-0.75, -0.57] % [Table 21, Figure 2], the efficacy of monotherapy has been demonstrated.

PMDA considers as follows:

In the Japanese phase III study (Study D1692C00006) evaluating monotherapy, the superiority of dapagliflozin at doses of 5 and 10 mg over placebo in the change in HbA1c from baseline to Week 24 of treatment (the primary endpoint) was demonstrated [Table 18]. In addition, the Japanese phase III long-term treatment study (Study D1692C00012) demonstrated persistence of efficacy for up to Week 52 in terms of the change from baseline in HbA1c in the monotherapy group [Table 21, Figure 2]. Based on the foregoing, the efficacy of monotherapy has been demonstrated.

4.(iii).B.(2).2 Efficacy of concomitant therapies

PMDA asked the applicant to explain the reason for the variability in the change in HbA1c among the concomitant therapy groups observed in the Japanese phase III long-term treatment study (Study D1692C00012).

The applicant responded as follows:

In this study, the change in HbA1c from baseline to Week 52 of treatment in the dapagliflozin + GLP-1 group was smaller than those in the other concomitant therapy groups [Table 21]. An investigation of its reason revealed that the duration of type 2 diabetes mellitus (mean ± SD) in subjects enrolled in the dapagliflozin + GLP-1 group was 13.4 ± 8.9 years, that was longer than those in the other concomitant therapy groups (7.2 ± 5.7 years in the dapagliflozin + SU group; 7.6 ± 6.2 years in the dapagliflozin + DPP-4 group; 4.9 ± 5.0 years in the dapagliflozin + α-GI group; 5.3 ± 3.9 years in the dapagliflozin + BG group; 5.5 ± 4.3 years in the dapagliflozin + TZD group; 5.8 ± 5.1 years in the dapagliflozin + glinide group). In addition, the proportion of subjects with baseline eGFR <60 was found to be higher in the dapagliflozin + GLP-1 group (32.0%) than in the other concomitant therapy groups (20.5% in the dapagliflozin + SU group; 21.0% in the dapagliflozin + DPP-4 group; 26.2% in the dapagliflozin + α-GI group; 7.0% in the dapagliflozin + BG group; 18.8% in the dapagliflozin + TZD group; 20.4% in the dapagliflozin + glinide group). The above differences were considered to have led to the smaller change in HbA1c in the dapagliflozin + GLP-1 group than those in the other concomitant therapy groups. Furthermore, the proportion of subjects who received hyperglycaemia rescue therapy in the dapagliflozin + GLP-1 group (32.0%) was higher than those in the other concomitant therapy groups (14.8% in the dapagliflozin + SU group; 11.3% in the dapagliflozin + DPP-4 group; 7.2% in the dapagliflozin + BG group; 3.1% in the dapagliflozin + TZD group; 2.0% in the dapagliflozin + glinide group; 0% in the dapagliflozin + α-GI group; 1.6% in the monotherapy group), supporting the applicant's discussion.

PMDA considers that the efficacy of each concomitant therapy has been confirmed even though the variability in the change in HbA1c was observed among the concomitant therapy groups in Study D1692C00012.

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

PMDA asked the applicant to explain the impact of a concomitant antidiabetic drug on the safety depending on the dose and type.

The applicant responded as follows:

In Study D1692C00012, the incidence of adverse events by dose level of glimepiride in the dapagliflozin + SU group was 71.6% (68 of 95 subjects) at ≤ 2 mg/day and 77.8% (21 of 27 subjects) at > 2 mg/day, showing no substantial difference according to dose level. The incidence of hypoglycaemia was 5.3% (5 of 95 subjects) at glimepiride ≤ 2 mg/day and 11.1% (3 of 27 subjects) at glimepiride > 2 mg/day, showing a higher incidence at > 2 mg/day, but no severe hypoglycaemia were reported. The incidence of adverse events by dose level of sitagliptin in the dapagliflozin + DPP-4 group was 72.9% (35 of 48 subjects) at ≤ 50 mg/day and 85.7% (12 of 14 subjects) at > 50 mg/day, showing a higher incidence at > 50 mg/day, but no events occurred with a particularly high incidence. The incidence of adverse events by type of antidiabetic drug in the dapagliflozin + α -GI group was 66.7% (2 of 3 subjects) for acarbose, 69.2% (18 of 26 subjects) for miglitol, and 59.4% (19 of 32 subjects) for voglibose. In the voglibose group, which had the largest number of subjects, the incidence by dose level of voglibose was 62.5% (10 of 16 subjects) at 0.6 mg/day and 56.3% (9 of 16 subjects) at 0.9 mg/day, showing no substantial difference according to dose. The incidence of adverse events by dose level of metformin in the dapagliflozin + BG group was 78.6% (44 of 56 subjects) at ≤ 750 mg/day and 80.0% (12 of 15 subjects) at > 750 mg/day, showing no substantial difference according to dose. The incidence of adverse events by dose level of pioglitazone in the dapagliflozin + TZD group was 68.8% (22 of 32 subjects) at ≤ 15 mg/day and 71.9% (23 of 32 subjects) at > 15 mg/day, showing no substantial difference according to dose. The adverse event related to oedema was face oedema (mild) alone reported by 1 subject (69-year-old woman) who received pioglitazone at 30 mg/day. The incidence of adverse events by type of antidiabetic drug in the dapagliflozin + glinide group was 59.1% (13 of 22 subjects) for mitiglinide and 77.8% (21 of 27 subjects) for nateglinide, showing a trend toward higher incidence in use of nateglinide. The incidence of adverse events by dose level of liraglutide in the dapagliflozin + GLP-1 group was 85.7% (6 of 7 subjects) at < 0.9 mg/day and 72.1% (31 of 43 subjects) at 0.9 mg/day, showing a higher incidence at < 0.9 mg/day, but evaluation was difficult because of the limited number of subjects receiving liraglutide at < 0.9 mg/day.

Regarding concomitant therapy with insulin, the incidence of adverse events (excluding hypoglycaemia) during 12 weeks of treatment in Foreign Study MB102009¹⁵⁰ was 65.2% (15 of 23 subjects) in the placebo group, 75.0% (18 of 24 subjects) in the dapagliflozin 10 mg group, and 66.7% (16 of 24 subjects) in the dapagliflozin 20 mg group, showing no substantial difference. The incidence of adverse events during 104 weeks of treatment in Foreign Study D1690C00006¹⁵¹ was 78.2% (154 of 197 subjects) in the placebo group, 80.2% (162 of 202 subjects) in the dapagliflozin 2.5 mg group, 78.3% (166 of 212 subjects) in the 5/10 mg group, and 80.1% (157 of 196 subjects) in the 10 mg group, showing no substantial difference. In Foreign Study MB102009, as oedema-related events, 2 events of oedema peripheral were reported by 2 subjects in the dapagliflozin 20 mg group (both subjects received concomitant TZD), but both were mild or moderate in severity and a causal relationship to the study drug was ruled out for the events. The incidence of oedema in Foreign Study D1690C00006 was 9.1% (18 of 197 subjects) in the placebo group, 5.0% (10 of 202 subjects) in the dapagliflozin 2.5 mg group, 3.3% (7 of 212 subjects) in the 5/10 mg group, and 5.6% (11 of 196 subjects) in the 10 mg group, showing a lower incidence in the dapagliflozin groups, and most events were mild or moderate in severity (severe oedema peripheral, 1 subject each in the placebo and dapagliflozin 2.5 mg groups).

¹⁵⁰ Foreign Study MB102009: A placebo-controlled, randomized, double-blind, parallel-group phase III study in patients with type 2 diabetes mellitus on insulin therapy (patients receiving continued treatment at a stable daily dose of metformin [at ≥ 1000 mg/day] and/or TZD [pioglitazone at ≥ 30 mg/day or rosiglitazone at 4 mg/day] for ≥ 6 weeks prior to study entry) that evaluated the efficacy and safety of dapagliflozin 10 and 20 mg in combination with insulin (treatment duration was 12 weeks).

¹⁵¹ Foreign Study D1690C00006: A placebo-controlled, randomized, double-blind, parallel-group phase III study in patients with type 2 diabetes mellitus on insulin therapy that evaluated the efficacy and safety of dapagliflozin 2.5, 5, and 10 mg in combination with insulin (consisting of a 24-week confirmatory study and the subsequent long-term extension periods [24-week + 56-week], for a total duration of 104 weeks). The group in which the dose level of dapagliflozin was switched from 5 mg to 10 mg at Week 49 of treatment is referred to as 5/10 mg group.

PMDA considers as follows:

Given the incidence of adverse events with monotherapy and each concomitant therapy, the safety of dapagliflozin is acceptable on the premise that appropriate caution statements are provided. Although there is no particular problem with the impact of concomitant antidiabetic drugs on the safety depending on their doses and types, given the limited number of subjects included in some studies on such potential impact, it is necessary to continue to collect information on the safety via post-marketing surveillance. PMDA further reviewed the following events of special safety interest.

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

The applicant explained as follows:

In Japanese Study D1692C00005, hypoglycaemia¹²⁹ were reported by 1 subject each in the placebo, dapagliflozin 2.5 mg, and dapagliflozin 10 mg groups, and among these, the event in the subject in the dapagliflozin 2.5 mg group was assessed as mild hypoglycaemia, and that in the subject in the 10 mg group was assessed as other hypoglycaemia. In Japanese Study D1692C00006, hypoglycaemia was reported by 2 subjects in the dapagliflozin 10 mg group (other hypoglycaemia). In Japanese Study D1692C00012, the incidence of hypoglycaemia¹²⁹ was 2.4% (6 of 249 subjects) in the monotherapy group; 6.6% (8 of 122 subjects) in the dapagliflozin + SU group; 3.2% (2 of 62 subjects) in the dapagliflozin + DPP-4 group; 0% (0 of 61 subjects) in the dapagliflozin + α-GI group; 2.8% (2 of 71 subjects) in the dapagliflozin + BG group; 1.6% (1 of 64 subjects) in the dapagliflozin + TZD group; 6.1% (3 of 49 subjects) in the dapagliflozin + glinide group; and 6.0% (3 of 50 subjects) in the dapagliflozin + GLP-1 group, showing a higher incidence in the dapagliflozin + SU, glinide, and GLP-1 groups, but no severe hypoglycaemia was reported.

In Foreign Study MB102009¹⁵⁰ evaluating concomitant therapy with insulin, the incidence of hypoglycaemia¹⁵² during 12 weeks of treatment was 8.7% (2 events in 2 of 23 subjects) in the placebo group; 8.3% (3 events in 2 of 24 subjects) in the dapagliflozin 10 mg group; and 16.7% (4 events in 4 of 24 subjects) in the 20 mg group. One event of severe hypoglycaemia was reported by 1 subject in the placebo group. In Foreign Study D1690C00006,¹⁵¹ the incidence of hypoglycaemia¹²⁹ during 24 weeks of treatment was 42.1% (487 events in 83 of 197 subjects) in the placebo group; 55.0% (808 events in 111 of 202 subjects) in the dapagliflozin 2.5 mg group; 47.6% (736 events in 101 of 212 subjects) in the 5 mg group; and 44.9% (468 events in 88 of 196 subjects) in the 10 mg group. The incidence of hypoglycaemia during 104 weeks of treatment was 61.9% (1451 events in 122 of 197 subjects) in the placebo group; 69.3% (2365 events in 140 of 202 subjects) in the dapagliflozin 2.5 mg group; 61.3% (2253 events in 130 of 212 subjects) in the 5/10 mg group; and 60.7% (1424 events in 119 of 196 subjects) in the 10 mg group. Most events of hypoglycaemia were classified as mild hypoglycaemia. The incidence of severe hypoglycaemia was 1.0% (3 events in 2 of 197 subjects) in the placebo group; 2.0% (13 events in 4 of 202 subjects) in the dapagliflozin 2.5 mg group; 1.4% (4 events in 3 of 212 subjects) in the 5/10 mg group; and 1.5% (3 events in 3 of 196 subjects) in the 10 mg group. Serious adverse events reported include hypoglycaemia in 2 subjects in the 5/10 mg group and hypoglycemic coma in 1 subject in the placebo group.

Updated data on a pooled analysis (30-MU¹⁵³) showed that the incidence of hypoglycaemia¹²⁹ in 30-MU (short-term) was 10.5% [242 of 2295 subjects] in the placebo group and 13.1% [309 of

¹⁵² Definition of hypoglycaemia: (a) Severe hypoglycaemia, Any symptomatic event with blood glucose <54 mg/dL that requires third-party assistance due to severe consciousness disturbed or behavior disorder and recovers immediately by treatment with glucose or glucagon; (b) Mild hypoglycaemia, Any symptomatic or asymptomatic event with blood glucose <54 mg/dL that does not meet the criteria of severe hypoglycaemia; (c) Other hypoglycaemia, Any event reported as an event suggestive of hypoglycaemia without any documentation of blood glucose levels.

¹⁵³ Data with cut-off date of █20█. The pooled data from all the clinical studies (30-MU [all studies]) consist of combined data from 5 phase IIb studies and 16 phase III studies (including Japanese Studies D1692C00005, D1692C00006, and D1692C00012),

2360 subjects] in the dapagliflozin 10 mg group, indicating a higher incidence in the dapagliflozin group.

Since the incidence of hypoglycaemia was higher in the dapagliflozin + SU, glinide, and GLP-1 groups in Japanese Study D1692C00012, PMDA asked the applicant to explain the safety of concomitant therapies with these drugs.

The applicant responded as follows:

Hypoglycaemia was observed in 8 subjects in the Dapagliflozin + SU group, including mild hypoglycaemia in 2 subjects (reported by 1 subject each receiving glimepiride at 1 and 3 mg/day) and other hypoglycaemia in 6 subjects, and no severe hypoglycaemia occurred. Among these 8 subjects, 4 subjects received glimepiride at 1 mg/day, 1 subject at 2 mg/day, and 3 subjects at 3 mg/day; thus, no events of hypoglycaemia were reported by subjects receiving glimepiride at \geq 4 mg/day. These 8 subjects all had baseline eGFR (mL/min/1.73 m²) of \geq 60 and $<$ 90 (mild renal impairment), but other background characteristics were similar between these subjects and subjects in the whole Dapagliflozin + SU group. In Foreign Study D1690C00005¹⁵⁴ evaluating glimepiride monotherapy combined with dapagliflozin, the incidence of hypoglycaemia¹²⁹ during 48 weeks of treatment was 6.8% (10 of 146 subjects) in the placebo group; 9.7% (15 of 154 subjects) in the dapagliflozin 2.5 mg group; 10.3% (15 of 145 subjects) in the dapagliflozin 5 mg group; and 11.3% (17 of 151 subjects) in the dapagliflozin 10 mg group, showing a trend toward higher incidence among subjects receiving dapagliflozin. Based on the above, caution statements will be included in the package insert regarding the need for paying attention to occurrence of hypoglycaemia and for considering dose reduction of SUs during the use of dapagliflozin + SUs. Of 3 subjects in the dapagliflozin + glinide group, 2 subjects (mild hypoglycaemia and other hypoglycaemia in 1 subject each) received nateglinide at 270 mg/day, and 1 subject (other hypoglycaemia) received mitiglinide at 30 mg/day. The 2 subjects receiving nateglinide at 270 mg/day had baseline eGFR of $<$ 60; thus, hypoglycaemia may have been related to the subjects' renal function. All of the 3 subjects who experienced hypoglycaemia (mild hypoglycaemia) in the dapagliflozin + GLP-1 group concomitantly used liraglutide at 0.9 mg/day, and 1 subject among these had eGFR $<$ 60.

Since the results of Foreign Study MB102066¹⁵⁵ evaluating the effect of renal glycosuria suggested that glucose is excreted in urine even in subjects showing low plasma glucose levels, PMDA asked the applicant to explain the possibility that dapagliflozin may induce protracted hypoglycaemia.

The applicant responded as follows:

The results of Foreign Study MB102066 showed that the adjusted geometric mean of the maximum tubular reabsorption capacity for glucose (T_{mG}) at baseline was higher in patients with type 2 diabetes mellitus (420 mg/min) by 32.2% than in healthy subjects (317 mg/min), with geometric mean ratio (patients with type 2 diabetes mellitus/healthy subjects) [two-sided 90% CI] of 1.322 [1.059, 1.650], suggesting that baseline T_{mG} was higher in patients with type 2 diabetes mellitus than in healthy subjects. The plasma glucose thresholds above which glucose was

and the pooled data from placebo-controlled studies consist of combined data from 13 studies including 3 phase IIb studies and 10 phase III studies. The pooled data from short-term placebo-controlled studies (30-MU [short-term]) consist of combined data from 13 short-term placebo-controlled studies (for \leq 24 weeks) (including Japanese Study D1692C00005), and the pooled data from short-term + long-term placebo-controlled studies (30-MU [short-term + long-term]) consist of combined data from short-term + long-term treatment periods (for \geq 48 and \leq 104 weeks) of 9 placebo-controlled studies.

¹⁵⁴ Foreign Study D1690C00005: A placebo-controlled, randomized, double-blind, parallel-group phase III study in patients with type 2 diabetes mellitus on monotherapy with SU at no less than half of the maximum recommended dose for \geq 8 weeks prior to study entry that evaluated the efficacy and safety of dapagliflozin 2.5, 5, and 10 mg in combination with glimepiride (24-week confirmatory study + 24-week extension period).

¹⁵⁵ Foreign Study MB102066: A study in healthy subjects and patients with type 2 diabetes mellitus aged \geq 18 and \leq 65 years that evaluated the maximum tubular reabsorption capacity for glucose (T_{mG}) and the plasma glucose threshold above which glucose appears in urine during multiple administration of dapagliflozin 10 mg for 7 days.

detected in urine in healthy subjects and patients with type 2 diabetes mellitus were 171 and 196 mg/dL, respectively, at baseline, but were decreased to ≤100 mg/dL by multiple administration of dapagliflozin for 7 days (37 and 21 mg/dL on Day 7, respectively). Thus, glucose may be excreted in urine even in subjects showing plasma glucose levels of <100 mg/dL. However, the filtered glucose would not be excreted completely because of glucose reabsorption in the kidneys. It is reported that, at a plasma glucose level of approximately 100 mg/dL, a total of <20% of the filtered glucose is excreted in urine.¹⁵⁶ Although no specific evaluation was performed, given the relationship between plasma glucose level and amount of filtered glucose detected in urine, the levels of glucose excreted in urine are expected to be low at plasma glucose levels of <100 mg/dL. A steady state is reached at Day 7 of treatment, and similar results are expected after long-term treatment. Urinary glucose excretion is related to the glucose reabsorption, filtered load of glucose (plasma glucose level × eGFR), and glucose reabsorption capacity. Once the plasma glucose levels are elevated beyond the glucose reabsorption capacity of SGLT2 and SGLT1, an amount of glucose is excreted in urine depending on the filtered load. Inhibition of SGLT2 by dapagliflozin prohibits glucose transport from the proximal tubules to circulating blood by SGLT2, leading to a substantial decrease in the glucose reabsorption capacity. The filtered load is proportional to the glucose reabsorption, and low plasma glucose levels lead to a reduced urinary glucose excretion even on treatment with dapagliflozin. Glucose is considered to be reabsorbed also by SGLT1. A review of the duration of severe hypoglycaemia observed in 30-MU (short-term + long-term), including data collected after hyperglycaemia rescue therapy, showed that the incidence of severe hypoglycaemia was 0.3% both in the dapagliflozin 10 mg (6 of 2026 subjects) and placebo (5 of 1956 subjects) groups. Severe hypoglycaemia was observed in 6 subjects in the dapagliflozin 10 mg group and the duration was 30 minutes to 3 hours. The patient with the longest duration of hypoglycaemia (3 hours) concomitantly used gliclazide (SU with half-life of 11 hours). Severe hypoglycaemia was observed in 5 subjects in the placebo group and the duration was 4 minutes to 2 hours 45 minutes; thus, there was no substantial difference from the dapagliflozin group.

Based on the above, dapagliflozin is unlikely to induce protracted hypoglycaemia.

PMDA accepted the applicant's response. However, given the findings including the trend toward a higher incidence of hypoglycaemia in the dapagliflozin + SU, glinide, and GLP-1 groups than in the monotherapy group observed in a Japanese clinical study, and the trend toward a higher incidence of hypoglycaemia in the dapagliflozin group than in the placebo group among subjects receiving concomitant insulin in the foreign clinical studies, it is necessary to provide an appropriate caution statement about hypoglycaemia, and to continue to collect information on hypoglycaemia via post-marketing surveillance.

4.(iii).B.(3).2 Polyuria/pollakiuria-related events

The applicant explained as follows:

In Japanese Study D1692C00005, pollakiuria was reported by 1 subject in the dapagliflozin 10 mg group, but no polyuria was reported. In Japanese Study D1692C00006, the incidence of pollakiuria was 1.1% (1 of 87 subjects) in the placebo group, 2.3% (2 of 86 subjects) in the dapagliflozin 5 mg group, and 4.5% (4 of 88 subjects) in the 10 mg group, showing a higher incidence in the dapagliflozin groups. Polyuria was reported only by 1 subject in the dapagliflozin 10 mg group. All events of polyuria and pollakiuria were assessed as adverse drug reactions. In Study D1692C00012, the incidence of pollakiuria was 5.2% (13 of 249 subjects) in the monotherapy group; 1.6% (2 of 122 subjects) in the dapagliflozin + SU group; 1.6% (1 of 62 subjects) in the dapagliflozin + DPP-4 group; 3.3% (2 of 61 subjects) in the dapagliflozin + α-GI group; 4.2% (3 of 71 subjects) in the dapagliflozin + BG group; 4.7% (3 of 64 subjects) in the dapagliflozin + TZD group; and 4.1% (2 of 49 subjects) in the dapagliflozin + glinide group. No

¹⁵⁶ Defronzo RA, et al. *Diabetes Care*. 2013;36:3169-76.

events of pollakiuria were reported in the dapagliflozin + GLP-1 group. All events except that reported by 1 subject in the dapagliflozin + α-GI group were assessed as adverse drug reactions. No adverse events of polyuria were reported.

Updated data on a pooled analysis (30-MU¹⁵³) showed that the incidence of pollakiuria-related adverse events (pollakiuria, polyuria, urine output increased) in 30-MU (short-term) was 1.2% (27 of 2295 subjects) in the placebo group and 3.3% (78 of 2360 subjects) in the dapagliflozin 10 mg group; the incidence of pollakiuria in 30-MU (short-term + long-term) was 1.4% (28 of 1956 subjects) in the placebo group and 3.9% (79 of 2026 subjects) in the dapagliflozin 10 mg group, showing a higher incidence in the dapagliflozin group in both pooled populations, but no serious events were reported.

Since there was a trend toward a higher incidence of polyuria/pollakiuria-related adverse events in the dapagliflozin group, PMDA considers that it is necessary to continue to collect information on polyuria/pollakiuria-related events via post-marketing surveillance.

4.(iii).B.(3).3 Urinary tract infections

The applicant explained as follows:

In Japanese Study D1692C00005, events suggestive of urinary tract infection were reported by 1 subject in the placebo group (cystitis [mild] in a female subject), 1 subject in the dapagliflozin 1 mg group (cystitis bacterial [mild] in a male subject), 1 subject in the 5 mg group (cystitis [moderate] in a female subject), and 2 subjects in the 10 mg group (cystitis [moderate] in a female subject, cystitis [mild] in a male subject). In Japanese Study D1692C00006, urinary tract infection-related events¹⁵⁷ were reported by 2 female subjects in the placebo group (cystitis [mild in both subjects]) and 2 female subjects in the dapagliflozin 10 mg group (cystitis [mild], urinary tract infection [moderate]), but no events were serious, and all events except that reported by 1 subject in the placebo group resolved with intervention; no recurrence of urinary tract infection-related events was reported by any subject. Regarding the time of onset, the urinary tract infection-related events occurred during Week 1 to 4 in 1 subject in the dapagliflozin 10 mg group and Week 17 to 20 in the other subject in the 10 mg group. No kidney infection (pyelonephritis) was found. In Japanese Study D1692C00012, the incidence of urinary tract infection-related events was 3.6% (9 of 249 subjects) in the monotherapy group; 3.3% (4 of 122 subjects) in the dapagliflozin + SU group; 0% (0 of 62 subjects) in the dapagliflozin + DPP-4 group; 1.6% (1 of 61 subjects) in the dapagliflozin + α-GI group; 2.8% (2 of 71 subjects) in the dapagliflozin + BG group; 3.1% (2 of 64 subjects) in the dapagliflozin + TZD group; 0% (0 of 49 subjects) in the dapagliflozin + glinide group; and 4.0% (2 of 50 subjects) in the dapagliflozin + GLP-1 group, showing no substantial difference. Seventeen of 20 subjects who experienced urinary tract infection-related events were female. Moderate urinary tract infection-related events included prostatitis in 1 subject in the Dapagliflozin + SU group and cystitis in 1 subject in the dapagliflozin + GLP-1 group, and no severe or serious events were observed. The time of onset of urinary tract infection-related events (including recurrences of the same event) with the number of events was Weeks 1 to 12 (9 events), Weeks 13 to 24 (5 events), Weeks 25 to 36 (4 events), Weeks 37 to 48 (3 events), and Weeks ≥49 (4 events), showing a higher frequency during Weeks 1 to 12, but a longer treatment duration was not associated with a higher risk of these events. No kidney infection was reported during 52 weeks of treatment.

In the concomitant use with insulin, 1 event of urinary tract infection (mild) was reported by 1 subject in the dapagliflozin 20 mg group in Foreign Study MB102009.¹⁵⁰ In Foreign Study D1690C00006,¹⁵¹ the incidence of events suggestive of urinary tract infection during 104 weeks of treatment was 5.6% (11 of 197 subjects) in the placebo group; 8.4% (17 of 202 subjects) in the

¹⁵⁷ Collected based on the list of preferred terms predefined by the applicant. Among events suggesting urinary tract infection, only those events that represented urinary tract infection were considered as urinary tract infection-related events.

dapagliflozin 2.5 mg group; 13.2% (28 of 212 subjects) in the 5/10 mg group; and 13.8% (27 of 196 subjects) in the 10 mg group, showing higher incidences in the dapagliflozin groups. During 104 weeks of treatment, severe urinary tract infection-related events were reported by 1 subject in the dapagliflozin 2.5 mg group (urinary tract infection) and 1 subject in the 5/10 mg group (pyelonephritis), but no serious events were reported. The incidences in female and male subjects during 104 weeks of treatment were 7.1% (7 of 99 subjects) and 4.1% (4 of 98 subjects), respectively, in the placebo group; 9.8% (10 of 102 subjects) and 7.0% (7 of 100 subjects), respectively, in the dapagliflozin 2.5 mg group; 18.8% (21 of 112 subjects) and 7.0% (7 of 100 subjects), respectively, in the dapagliflozin 5/10 mg group; and 19.4% (21 of 108 subjects) and 6.8% (6 of 88 subjects), respectively, in the dapagliflozin 10 mg group; showing higher incidences among female subjects and higher incidences in dapagliflozin groups than in the placebo group.

Updated data on a pooled analysis (30-MU¹⁵³) showed that the incidence of urinary tract infection-related events in 30-MU (short-term) was 3.5% (88 events in 81 of 2295 subjects) in the placebo group and 4.7% (132 events in 110 of 2360 subjects) in the dapagliflozin 10 mg group, showing a higher incidence in the dapagliflozin group. The incidences in female and male subjects were 6.7% (64 of 952 subjects) and 1.3% (17 of 1343 subjects), respectively, in the placebo group and 8.5% (85 of 1003 subjects) and 1.8% (25 of 1357 subjects), respectively, in the dapagliflozin 10 mg group, showing higher incidences among female subjects. The majority of events were mild or moderate in severity. In both groups, many ($\geq 80\%$) of the subjects who experienced a urinary tract infection-related event received intervention against it. Additional intervention was required for 10 of 88 events (11.4%) in the placebo group and 15 of 132 events (11.4%) in the dapagliflozin group. The proportion of subjects who experienced a urinary tract infection-related event in the placebo and dapagliflozin groups by frequency was 91.4% (74 of 81 subjects) and 83.6% (92 of 110 subjects), respectively, for once; 8.6% (7 of 81 subjects) and 12.7% (14 of 110 subjects), respectively, for twice; 0% and 3.6% (4 of 110 subjects), respectively, for three times; no subjects experienced the event 4 times or more. Twenty-six of 110 subjects (23.6%) who experienced urinary tract infection-related event(s) in the dapagliflozin 10 mg group had a prior history of urinary tract infection-related conditions (nocturia, recurrent urinary tract infection, benign prostatic hyperplasia, renal and urinary calculus); similarly, 24 of 81 subjects (29.6%) who experienced urinary tract infection-related event(s) in the placebo group had a prior history of the related conditions. Recurrent urinary tract infection was defined as urinary tract infection that occurred at least twice within 6 months or at least 3 times within a year regardless of when they occurred, and the proportion of subjects who had a prior history of recurrent urinary tract infection among subjects who experienced a urinary tract infection-related event was determined. The results revealed that the proportion was 18.5% (15 of 81 subjects) in the placebo group and 10.9% (12 of 110 subjects) in the dapagliflozin 10 mg group. No differences were observed in severity or duration of the urinary tract infection or type of intervention between subjects with and without a prior history of urinary tract infection, and most of the infections resolved with 1 course of standard treatment.

The incidence of urinary tract infection-related events in 30-MU (short-term + long-term) was 6.2% (121 of 1956 subjects) in the placebo group and 8.6% (174 of 2026 subjects) in the dapagliflozin 10 mg group, showing a higher incidence in the dapagliflozin group.

The incidence of urinary tract infection-related events was evaluated according to baseline HbA1c and BMI using data of 30-MU (short-term). The incidences by baseline HbA1c in the placebo and dapagliflozin 10 mg groups were 4.0% (6 of 151 subjects) and 2.2% (3 of 137 subjects), respectively, for subjects with HbA1c of <7%; 3.1% (32 of 1025 subjects) and 4.6% (45 of 977 subjects), respectively, for subjects with HbA1c of $\geq 7\%$ and <8%; and 4.0% (55 of 1388 subjects) and 5.0% (62 of 1246 subjects), respectively, for subjects with HbA1c of $\geq 8\%$. The incidence was slightly higher in the dapagliflozin group than in the placebo group except among subjects with baseline HbA1c of <7%. The incidence of urinary tract infection-related events did not vary

depending on baseline HbA1c among subjects in the placebo group, while the incidence increased with increasing HbA1c among subjects in the dapagliflozin group. The incidences by baseline BMI in the placebo and dapagliflozin 10 mg groups were 4.1% (10 of 243 subjects) and 4.0% (7 of 173 subjects), respectively, for subjects with BMI of <25 kg/m²; 2.6% (20 of 759 subjects) and 4.4% (31 of 709 subjects), respectively, for subjects with BMI of ≥25 and <30 kg/m²; and 4.0% (63 of 1562 subjects) and 4.9% (72 of 1478 subjects), respectively, for subjects with BMI of ≥30 kg/m². The incidence was slightly higher in the dapagliflozin group than in the placebo group except among subjects with BMI <25 kg/m². There was no consistent trend in incidence of urinary tract infection-related events according to baseline BMI among subjects in the placebo group, while a trend was observed for the incidence to increase with increasing BMI among subjects in the dapagliflozin group.

The incidence of kidney infection (including pyelonephritis) in 30-MU (all studies) was 0.2% (8 events in 7 of 3403 subjects) in the control group and 0.1% (7 events in 7 of 5936 subjects) in the dapagliflozin group, showing low incidences with no substantial difference. The incidence rate was 0.002/person years in the control group and 0.001/person years in the dapagliflozin group. Events reported by 1 subject in the dapagliflozin group (urosepsis) and 4 subjects in the control group (pyelonephritis in 2 subjects, cystitis/pyelonephritis and nephrolithiasis/pyelonephritis in 1 subject each) were assessed as serious adverse events. Based on the above, the risk of dapagliflozin-related exacerbation of kidney infection is considered to be limited. In the dapagliflozin group, septic shock in 6 subjects and sepsis in 4 subjects were observed (these events were not observed in the control group). A causal relationship to the study drug was ruled out for septic shock reported by the 6 subjects (4 male and 2 female foreign subjects aged 50-69 years). These 6 subjects all had co-morbid condition(s) in addition to diabetes mellitus, and included 4 subjects with diabetic complication(s). There was no clear trend in the type of organs affected by septic shock. The causes of these events included cholecystitis (2 subjects), pneumonia (1 subject), a complication of pneumonia and urinary tract infection (1 subject), and a complication of diabetic foot infection and extensive enterocolitis leading to colectomy and ileostomy (1 subject), all of which have been known as infections that could easily lead to sepsis due to bacteraemia (The Japanese Guidelines for the Management of Sepsis). These infections were considered to be the causes of sepsis in 3 of these subjects, including 1 subject who experienced sepsis after an event of gastrointestinal haemorrhage and 2 subjects in whom sepsis led to study treatment discontinuation. The interval between the start of treatment and the onset of septic shock in the 6 subjects was in the range of 33 to 1238 days. The 4 subjects who experienced sepsis (3 male and 1 female foreign subjects aged 68-73 years) all had co-morbid condition(s) in addition to diabetes mellitus, and included 3 subjects with diabetic complication(s). These subjects included 1 subject with culture-negative sepsis, 1 subject with sepsis related to the most recent biopsy prostate, and 1 subject with sepsis due to a respiratory infection associated with very severe pneumonia. The only event considered related to urinary tract infection was reported by 1 subject who had a permanent indwelling bladder catheter for a long time; and the only event assessed as an adverse drug reaction was also reported by this subject. The interval between the start of treatment and the onset of sepsis in the 4 subjects was in the range of 135 to 562 days. As described above, septic shock or sepsis was reported by vulnerable subjects susceptible to infection, most of whom had co-morbid condition(s) or diabetic complication(s) in addition to diabetes mellitus. There was no consistent trend in the time to event onset or the type of putative first infected organs, with no consistent outcomes.

Given the findings including the trend toward a higher incidence of urinary tract infection-related events in the dapagliflozin group than in the placebo group, the reports of sepsis by subjects receiving dapagliflozin in the clinical studies, and the limited number of subjects included in the clinical studies, PMDA considered that it is necessary to provide an appropriate caution statement about urinary tract infections, and to continue to collect information on urinary tract infections (also including pyelonephritis and sepsis) via post-marketing surveillance.

4.(iii).B.(3).4 Genital infections

The applicant explained as follows:

In Japanese Study D1692C00005, events suggestive of genital infection were reported by 1 subject in the dapagliflozin 2.5 mg group (pruritus genital [mild] in a female subject) and 1 subject in the dapagliflozin 5 mg group (balanitis [mild] in a male subject). In Japanese Study D1692C00006, genital infection-related events¹⁵⁸ were reported by 1 subject in the dapagliflozin 5 mg group (vulvovaginal candidiasis [mild] in a female subject) and 1 subject in the 10 mg group (vulvitis [mild] in a female subject), but there were neither serious adverse events nor adverse events leading to treatment discontinuation, with no recurrence of genital infection-related events reported. The events of vulvitis observed in the dapagliflozin 10 mg group resolved without intervention. In Japanese Study D1692C00012, the incidence of genital infection-related events was 2.8% (7 of 249 subjects) in the monotherapy group; 2.5% (3 of 122 subjects) in the dapagliflozin + SU group; 3.2% (2 of 62 subjects) in the dapagliflozin + DPP-4 group; 1.6% (1 of 61 subjects) in the dapagliflozin + α-GI group; 2.8% (2 of 71 subjects) in the dapagliflozin + BG group; 0% (0 of 64 subjects) in the dapagliflozin + TZD group; 4.1% (2 of 49 subjects) in the dapagliflozin + glinide group; and 4.0% (2 of 50 subjects) in the dapagliflozin + GLP-1 group, showing no substantial difference. A moderate genital infection-related event was reported only by 1 subject in the monotherapy group (vulvovaginal candidiasis/vaginitis bacterial). No genital infection-related events were assessed as serious, but 1 subject (vulvovaginal pruritus) in the Dapagliflozin + DPP-4 group discontinued treatment. The time of onset of genital infection-related events (including recurrences of the same event) was Weeks 1 to 12 for 14 events, Weeks 13 to 24 for 3 events, Weeks 25 to 36 for 2 events, Weeks 37 to 48 for 1 event, and Weeks ≥49 for 1 event, showing a higher frequency during Weeks 1 to 12, but a longer treatment duration was not associated with a higher risk of these events.

In the concomitant use with insulin, the incidence of genital infection during 12 weeks of treatment in Foreign Study MB102009¹⁵⁰ was 4.3% (1 of 23 subjects) in the placebo group and 20.8% (5 of 24 subjects) in the dapagliflozin 20 mg group, showing a higher incidence in the dapagliflozin 20 mg group; no events of genital infection were reported by subjects in the 10 mg group. All events were mild or moderate in severity and no serious adverse events were reported. In Foreign Study D1690C00006,¹⁵¹ the incidence of events suggestive of genital infection during 104 weeks of treatment was 3.0% (6 of 197 subjects) in the placebo group, 7.4% (15 of 202 subjects) in the dapagliflozin 2.5 mg group, 12.7% (27 of 212 subjects) in the 5/10 mg group, and 14.3% (28 of 196 subjects) in the 10 mg group, showing a higher incidence in the dapagliflozin groups. Events suggestive of severe genital infection were reported by 1 subject in the placebo group (pruritus genital), 1 subject in the dapagliflozin 2.5 mg group (genital infection fungal), 1 subject in the 5/10 mg group (balanitis), and 1 subject in the 10 mg group (vulvovaginal mycotic infection), but no serious events were reported. The incidence in female and male subjects during 104 weeks of treatment was 6.1% (6 of 99 subjects) and 0% (0 of 98 subjects), respectively, in the placebo group; 9.8% (10 of 102 subjects) and 5.0% (5 of 100 subjects), respectively, in the dapagliflozin 2.5 mg group; 21.4% (24 of 112 subjects) and 3.0% (3 of 100 subjects), respectively, in the dapagliflozin 5/10 mg group; and 14.8% (16 of 108 subjects) and 13.6% (12 of 88 subjects), respectively, in the dapagliflozin 10 mg group; showing a higher incidence in female subjects and a higher incidence in dapagliflozin groups than in the placebo group.

Updated data from a pooled analysis (30-MU¹⁵³) showed that the incidence of genital infection-related events in 30-MU (short-term) was 0.6% (15 events in 14 of 2295 subjects) in the placebo group and 5.5% (154 events in 130 of 2360 subjects) in the dapagliflozin 10 mg group, showing a higher incidence in the dapagliflozin 10 mg group. The incidence in female and male subjects

¹⁵⁸ Collected based on the list of preferred terms predefined by the applicant. Among events suggesting genital infection, only those events that presented genital infection were considered as genital infection-related events.

was 1.2% (11 of 952 subjects) and 0.2% (3 of 1343 subjects), respectively, in the placebo group and 8.4% (84 of 1003 subjects) and 3.4% (46 of 1357 subjects), respectively, in the dapagliflozin 10 mg group, showing a higher incidence in female subjects. The majority of events were mild or moderate in severity. The events with a high incidence were vulvovaginal mycotic infection (0.7% in the placebo group, 3.4% in the dapagliflozin 10 mg group) in female subjects and balanitis (0% in the placebo group, 2.1% in the dapagliflozin 10 mg group) in male subjects. In both groups, many ($\geq 80\%$) of the subjects who experienced a genital infection-related event received intervention against it. Additional intervention was required for 0 of 15 events (0%) in the placebo group and 9 of 154 events (5.8%) in the dapagliflozin 10 mg group. The proportion of subjects who experienced a genital infection-related event only once was 92.9% (13 of 14 subjects) in the placebo group and 83.1% (108 of 130 subjects) in the dapagliflozin 10 mg group, the proportion of those who experienced twice was 7.1% (1 of 14 subjects) in the placebo group and 15.4% (20 of 130 subjects) in the dapagliflozin 10 mg group, and the proportion of those who experienced 3 times was 0% in the placebo group and 1.5% (2 of 130 subjects) in the dapagliflozin 10 mg group; no subjects experienced the event 4 times or more. Recurrent genital yeast infection was defined as genital infection that occurred prior to enrollment in the study at least 3 times within 6 months regardless of when they occurred and the proportion of subjects who had a prior history of recurrent genital yeast infection among subjects who experienced a genital infection-related events was determined. The results revealed that the proportion was 14.3% (2 of 14 subjects) in the placebo group and 6.2% (8 of 130 subjects) in the dapagliflozin 10 mg group. No differences were observed in severity or duration of the genital infection or type of the intervention between subjects with and without a prior history of genital infection, and most of the infections resolved with 1 course of standard treatment.

The incidence of genital infection-related events in 30-MU (short-term + long-term) was 1.0% (19 of 1956 subjects) in the placebo group and 7.7% (156 of 2026 subjects) in the dapagliflozin 10 mg group, showing a higher incidence in the dapagliflozin 10 mg group.

The incidence of genital infection-related events was evaluated according to baseline HbA1c and BMI using data of 30-MU (short-term). The incidences by baseline HbA1c were 0% (0 of 151 subjects) in the placebo group and 2.9% (4 of 137 subjects) in the dapagliflozin 10 mg group (subjects with HbA1c of $<7\%$); 0.8% (8 of 1025 subjects) in the placebo group and 6.0% (59 of 977 subjects) in the dapagliflozin 10 mg group (subjects with HbA1c of $\geq 7\%$ and $<8\%$); and 0.6% (9 of 1388 subjects) in the placebo group and 5.4% (67 of 1246 subjects) in the dapagliflozin 10 mg group (subjects with HbA1c of $\geq 8\%$). The incidence of genital infection-related events did not depend on baseline HbA1c among subjects in the placebo group, while in the dapagliflozin 10 mg group, the incidence was lower among subjects with baseline HbA1c of $<7\%$ than among subjects with baseline HbA1c of $\geq 7\%$ and $<8\%$ or of $\geq 8\%$. The incidences by baseline BMI were 0.8% (2 of 243 subjects) in the placebo group and 1.2% (2 of 173 subjects) in the dapagliflozin 10 mg group (subjects with BMI $<25 \text{ kg/m}^2$); 0.9% (7 of 759 subjects) in the placebo group and 5.1% (36 of 709 subjects) in the dapagliflozin 10 mg group (subjects with BMI of ≥ 25 and $<30 \text{ kg/m}^2$); and 0.5% (8 of 1562 subjects) in the placebo group and 6.2% (92 of 1478 subjects) in the dapagliflozin 10 mg group (subjects with BMI $\geq 30 \text{ kg/m}^2$). There was no difference in incidence of genital infection-related events according to baseline BMI among subjects in the placebo group, while a trend was observed for the incidence to be higher with increasing BMI among subjects in the dapagliflozin 10 mg group.

Given the trend toward a higher incidence of genital infection-related events in the dapagliflozin group than in the placebo group, PMDA considers that it is necessary to provide an appropriate caution statement about genital infections, and to continue to collect information on genital infections via post-marketing surveillance.

4.(iii).B.(3).5 Volume depletion

The applicant explained as follows:

Following multiple oral administration of dapagliflozin 10 mg for 7 days in foreign patients with type 2 diabetes mellitus in Foreign Study MB102057, the mean change from baseline in the daily urine volume was 823 mL on Day 1, decreased on Day 2 (137 mL), and remained at similar levels on Days 4 to 7 (243 mL on Day 4, 230 mL on Day 7). In Japanese Study D1692C00005, no events of hypotension, dehydration, and hypovolaemia were reported. Also in Japanese Study D1692C00006, no events related to volume depletion¹⁵⁹ were reported. In Japanese Study D1692C00012, the incidence of events related to volume depletion was 1.2% (3 of 249 subjects) in the monotherapy group, 1.6% (1 of 62 subjects) in the dapagliflozin + DPP-4 group, and 1.4% (1 of 71 subjects) in the dapagliflozin + BG group; no such events were reported in the other concomitant therapy groups. The event with the highest incidence was orthostatic hypotension and the incidence was 0.8% (2 of 249 subjects) in the monotherapy group and 1.4% (1 of 71 subjects) in the dapagliflozin + BG group.

Regarding effects on laboratory parameters, in Japanese Study D1692C00006, no clinically significant changes were observed in the placebo group, while in the dapagliflozin group, increases from baseline in haemoglobin, haematocrit, and red blood cell count were observed after 24 weeks of treatment and the parameters returned to baseline levels during the follow-up period. Notably abnormal values (haematocrit >55%, haemoglobin >18 g/dL) were observed in 1 subject in the dapagliflozin 5 mg group, but these were not associated with thromboembolism-related adverse events (e.g., transient ischaemic attack, stroke, venous thromboembolism). No clinically meaningful changes were observed in white blood cell count or platelet count. In Japanese Study D1692C00012, increases from baseline in haemoglobin, haematocrit, and red blood cell count were observed after 52 weeks of treatment that returned during the follow-up period. No substantial differences were observed across the concomitant therapy groups. The incidence of abnormal values of haematocrit (>55%) during 52 weeks of treatment was 1.2% (3 of 246 subjects) in the monotherapy group, 1.7% (2 of 120 subjects) in the dapagliflozin + SU group, 1.6% (1 of 61 subjects) in the dapagliflozin + α-GI group, and 4.0% (2 of 50 subjects) in the dapagliflozin + GLP-1 group; no such abnormalities were observed in the dapagliflozin + DPP-4, BG, TZD, and glinide groups. Abnormal values of haemoglobin (>18 g/dL) were reported by 9 subjects (1.3%), and the incidence was 1.6% (4 of 246 subjects) in the monotherapy group, 2.5% (3 of 120 subjects) in the dapagliflozin + SU group, 1.6% (1 of 61 subjects) in the dapagliflozin + α-GI group, and 2.0% (1 of 50 subjects) in the dapagliflozin + GLP-1 group; no such abnormalities were observed in the dapagliflozin + DPP-4, BG, TZD, and glinide groups. Thromboembolism-related adverse events associated with haematocrit high and haemoglobin high were not reported. No clinically meaningful changes were observed in white blood cell count or platelet count.

In the concomitant use with insulin, in Foreign Study MB102009,¹⁵⁰ 1 event of dehydration (severe) was reported by 1 subject in the dapagliflozin 10 mg group during 12 weeks of treatment. This subject was a 67-year-old male (baseline eGFR of 74 mL/min/1.73 m²) who received concomitant enalapril and furosemide, and experienced dehydration, blood creatinine increased, and blood urea increased (all were severe) on Day 8 and discontinued treatment with dapagliflozin on Day 11 (the change from baseline in eGFR at discontinuation was -35.12 mL/min/1.73 m²). This subject's condition was diagnosed as renal failure (severe), which was assessed as an adverse drug reaction, but the event (renal failure) resolved on Day 46. The incidence of events related to volume depletion during 104 weeks of treatment in Foreign Study D1690C00006¹⁵¹ was 1.0% (2 of 197 subjects) in the placebo group, 2.5% (5 of 202 subjects) in the dapagliflozin 2.5 mg group, 2.4% (5 of 212 subjects) in the 5/10 mg group, and 2.0% (4 of 196 subjects) in the 10 mg group, showing a higher incidence in the dapagliflozin groups. All events were mild or moderate in

¹⁵⁹ Events related to hypotension, hypovolaemia (LLT), and dehydration

severity. A serious event (syncope) was reported by 1 subject in the dapagliflozin 5/10 mg group, but a causal relationship to the study drug was ruled out for this event. Regarding effects on laboratory parameters, no haemoglobin or haematocrit abnormalities were reported in Foreign Study MB102009. In Foreign Study D1690C00006, haematocrit high (>55%) and haemoglobin high (>18 g/dL) were reported by 1 to 4 subjects in each dose group of dapagliflozin and by 1 subject in the placebo group.

Updated data from a pooled analysis (30-MU¹⁵³) showed that the incidence of events related to volume depletion in 30 -MU (short-term) was 0.7% (17 of 2295 subjects) in the placebo group and 1.1% (27 of 2360 subjects) in the dapagliflozin 10 mg group, showing a higher incidence in the dapagliflozin group [Table 25]. Approximately 20% of the events related to volume depletion (3 of 17 subjects in the placebo group, 5 of 27 subjects in the dapagliflozin 10 mg group) occurred within the first 2 weeks of treatment, about half of such events (9 of 17 subjects in the placebo group, 14 of 27 subjects in the dapagliflozin 10 mg group) occurred within the first 8 weeks of treatment, and approximately 80% of such events (13 of 17 subjects in the placebo group, 22 of 27 subjects in the dapagliflozin group) occurred within the first 14 weeks of treatment.

The incidence of events related to volume depletion in 30-MU (short-term + long-term) was 1.4% (27 of 1956 subjects) in the placebo group and 1.9% (38 of 2026 subjects) in the dapagliflozin 10 mg group [Table 25]. Approximately 10% of the events related to volume depletion (3 of 27 subjects in the placebo group, 4 of 38 subjects in the dapagliflozin 10 mg group) occurred within the first 2 weeks of treatment, approximately 30% of such events (9 of 27 subjects in the placebo group, 13 of 38 subjects in the dapagliflozin 10 mg group) occurred within the first 8 weeks of treatment, approximately 50% of such events (12 of 27 subjects in the placebo group, 21 of 38 subjects in the dapagliflozin group) occurred within the first 14 weeks of treatment, and approximately 70% of such events (18 of 27 subjects in the placebo group, 28 of 38 subjects in the dapagliflozin group) occurred within the first 30 weeks of treatment.

Table 25. Incidence of events related to volume depletion in 30-MU (pooled analysis)

Event term	30-MU (short-term)		30-MU (short-term + long-term)	
	Placebo group (n = 2295)	Dapagliflozin 10 mg group (n = 2360)	Placebo group (n = 1956)	Dapagliflozin 10 mg group (n = 2026)
Events related to volume depletion	17 (0.7)	27 (1.1)	27 (1.4)	38 (1.9)
Hypotension	5 (0.2)	15 (0.6)	6 (0.3)	18 (0.9)
Syncope	3 (0.1)	6 (0.3)	10 (0.5)	11 (0.5)
Dehydration	0 (0.0)	2 (0.1)	0 (0.0)	1 (0.0 ^a)
Orthostatic hypotension	6 (0.3)	2 (0.1)	7 (0.4)	3 (0.1)
Blood pressure decreased	1 (0.0 ^a)	1 (0.0 ^a)	2 (0.1)	2 (0.1)
Urine flow decreased	0 (0.0)	1 (0.0 ^a)	0 (0.0)	3 (0.1)
Circulatory collapse	1 (0.0 ^a)	0 (0.0)	2 (0.1)	1 (0.0 ^a)
Urine output decreased	1 (0.0 ^a)	0 (0.0)	1 (0.1)	1 (0.0 ^a)

Number of subjects with event (incidence %), MedDRA/J ver.15.1

a) One subject experienced this event, but the value is reported as 0.0 after rounding to 1 significant digit.

Regarding effects on laboratory parameters, the proportion of subjects with haematocrit high (>55%) was 0.4% (8 of 2295 subjects) in the placebo group and 1.3% (31 of 2360 subjects) in the dapagliflozin 10 mg group and the proportion of subjects with haemoglobin high (>18 g/dL) was 0.5% (11 of 2295 subjects) in the placebo group and 1.5% (36 of 2360 subjects) in the dapagliflozin 10 mg group, the proportions were higher in the dapagliflozin 10 mg group in 30-MU (short-term). Many of these notable laboratory abnormalities were observed singly. In addition, these notable laboratory abnormalities involving haemoglobin or haematocrit were generally not associated with adverse events. One subject each in the dapagliflozin 10 mg and

placebo groups (both in Foreign Study D1690C00019¹⁶⁰ conducted in subjects with type 2 diabetes mellitus who had a cardiovascular disease) experienced both haematocrit abnormally high and a thromboembolic event during the treatment period, but a causal relationship to the study drug was ruled out for these thromboembolic events. The mean change from baseline to Week 24 of treatment in haematocrit was greater in the dapagliflozin 10 mg group (2.3%) than in the placebo group (-0.3%). The mean change in haemoglobin was greater in the dapagliflozin 10 mg group (0.6 g/dL) than in the placebo group (-0.1 g/dL).

In 30-MU (short-term + long-term), the proportions of subjects with haematocrit high (>55%) were 0.6% (11 of 1956 subjects) in the placebo group and 2.1% (42 of 2026 subjects) in the dapagliflozin 10 mg group, and the proportions of subjects with haemoglobin high (>18 g/dL) were 0.7% (14 of 1956 subjects) in the placebo group and 2.2% (45 of 2026 subjects) in the dapagliflozin 10 mg group. The only additional subject who experienced both haematocrit abnormally high and a thromboembolic event during the treatment period other than those in 30-MU (short-term) was a subject in the dapagliflozin 5 mg group in Study D1690C00006¹⁵¹ evaluating combination therapy with insulin, but a causal relationship to the study drug was ruled out for the thromboembolic event.

PMDA asked the applicant to explain the safety of dapagliflozin including the safety of combination treatment with diuretics.

The applicant responded as follows:

In Japanese clinical studies, evaluation of the effects of concomitant diuretics on body fluid volume and/or electrolytes was difficult due to the limited number of subjects receiving concomitant diuretics, but neither consistent age-dependent changes nor clinically meaningful changes in body fluid volume-related laboratory parameters were observed.

In an analysis of the pooled population from placebo-controlled studies (short-term, short-term + long-term) according to the use or non-use of concomitant loop diuretics, thiazide diuretics, or angiotensin converting enzyme inhibitor and/or angiotensin receptor blockers (ACE-I/ARB) using a pooled data as of 2010 (SCS¹⁶¹), the incidence of events related to volume depletion across the overall population was 0.4% (5 of 1393 subjects) in the placebo group and 0.7% (24 of 3291 subjects) in the all dose groups of dapagliflozin, and the incidence in the population of subjects receiving concomitant loop diuretics was 1.8% (1 of 55 subjects) in the placebo group and 5.3% (6 of 114 subjects) in the all dose groups of dapagliflozin; there was a trend toward a difference between the overall population and population receiving concomitant loop diuretics, but the incidences of volume depletion-related events in the other subgroups were similar to that in the overall population.

The incidences of events related to volume depletion in 30-MU (short-term) according to the use or non-use of loop diuretics were 1.5% (4 of 267 subjects) in the placebo group and 2.5% (6 of 236 subjects) in the dapagliflozin 10 mg group among subjects receiving concomitant loop diuretics, and 0.6% (13 of 2028 subjects) in the placebo group and 1.0% (21 of 2124 subjects) in the dapagliflozin 10 mg group among subjects not receiving concomitant loop diuretics. The

¹⁶⁰ Foreign Study D1690C00019: A placebo-controlled, double-blind, comparative phase III study in patients with type 2 diabetes mellitus on antihypertensive treatment for cardiovascular disease that evaluated the efficacy and safety of 10 mg of supplemental dapagliflozin (consisting of 24-week confirmatory study and the subsequent long-term extension periods [28-week + 52-week], for a total duration of 104 weeks).

¹⁶¹ Pooled data of 14 studies from 3 phase IIb studies and 11 phase III studies, for which results were obtained prior to European regulatory submission in 2010. The pooled population from short-term placebo-controlled studies was based on combined short-term treatment data (for ≤24 weeks) from placebo controlled studies (a total of 12 studies including Japanese phase IIb study D1692C00005), and pooled population from short-term + long-term placebo-controlled studies was based on combined short-term + long-term treatment data (for ≥48 and ≤102 weeks) from placebo controlled studies (a total of 5 studies). Data from Studies MB102021 and MB102032, which were included in the pooled data submitted for regulatory approval in Europe, are not included in the pooled data of 30-MU, because dapagliflozin 10 mg group was not used in these studies.

incidences in 30-MU (short-term + long-term) were 2.7% (7 of 260 subjects) in the placebo group and 3.0% (7 of 234 subjects) in the dapagliflozin 10 mg group among subjects receiving concomitant loop diuretics, and 1.2% (20 of 1696 subjects) in the placebo group and 1.7% (31 of 1792 subjects) in the dapagliflozin 10 mg group among subjects not receiving concomitant loop diuretics. In 30-MU (short-term), the incidence was higher among subjects receiving concomitant loop diuretics than among subjects not receiving concomitant loop diuretics in both the placebo and dapagliflozin groups, and the incidence was higher in the dapagliflozin group than in the placebo group among both subjects receiving and not receiving concomitant loop diuretics.

An evaluation of other patient populations relatively susceptible to diuretic effect (elderly patients, patients with renal impairment) using data from 30-MU (short-term) revealed that the incidences of events related to volume depletion were 0.7% (11 of 1584 subjects) in the placebo group and 0.9% (16 of 1695 subjects) in the dapagliflozin 10 mg group in subjects aged <65 years, and 0.8% (6 of 711 subjects) in the placebo group and 1.7% (11 of 665 subjects) in the dapagliflozin 10 mg group in subjects aged ≥65 years, showing a higher incidence in subjects aged ≥65 years than in subjects aged <65 years as well as a higher incidence in the dapagliflozin group than in the placebo group. In terms of renal function, the incidences of events related to volume depletion were 1.5% (4 of 268 subjects) in the placebo group and 1.9% (5 of 265 subjects) in the dapagliflozin 10 mg group in subjects with baseline eGFR (mL/min/1.73 m²) of ≥30 and <60, and 0.6% (13 of 2025 subjects) in the placebo group and 1.1% (22 of 2094 subjects) in the dapagliflozin 10 mg group in subjects with baseline eGFR of ≥60, showing a higher incidence in patients with moderate renal impairment in both groups as well as a higher incidence in the dapagliflozin group than in the placebo group.

Based on the above, although dapagliflozin is unlikely to increase the risk of dehydration and its associated complications, a higher incidence of events related to volume depletion was observed in the dapagliflozin group than in the placebo group among subjects who received concomitant loop diuretics, who are ≥65 years of age, or who have moderate renal impairment; therefore, a caution statement will be included in the package insert regarding volume depletion. In addition, although the risk of notable hematologic abnormalities is considered to be limited, a careful follow-up/monitoring will be conducted in the future in light of the high haematocrit values observed in the dapagliflozin groups in Japanese and foreign clinical studies.

PMDA considers as follows:

The incidence of events related to volume depletion tended to be higher among elderly subjects, patients receiving concomitant drug(s) (e.g., diuretics), and patients with renal impairment, etc. In addition, although no relationship with thromboembolic events has been found at present, some subjects experienced an elevation in haematocrit, and the number of subjects and treatment duration in clinical studies were limited. Based on the above, it is necessary to provide an appropriate caution statement about volume depletion, and to continue to collect information on volume depletion (also including effects on haematocrit values) via post-marketing surveillance.

4.(iii).B.(3).6 Weight decreased

The applicant explained as follows:

The changes in body weight from baseline (adjusted mean ± SE) in Study D1690C00012¹⁶² evaluating effects of dapagliflozin on body weight and body composition were -1.24 ± 0.3126 kg (n = 86) in the placebo group and -3.35 ± 0.3194 kg (n = 83) in the dapagliflozin 10 mg group at Week 24; -1.89 ± 0.3898 kg (n = 84) in the placebo group and -4.24 ± 0.3999 kg (n = 81) in the dapagliflozin 10 mg group at Week 50; and -2.12 ± 0.4315 kg (n = 71) in the placebo group and

¹⁶² Foreign Study D1690C00012: A placebo-controlled, randomized, double-blind, parallel-group study in patients with type 2 diabetes mellitus (women aged ≥55 and ≤75 years who experienced menopause [or were hysterectomized] at least 5 years prior to informed consent or men aged ≥30 and ≤75 years) that evaluated the efficacy and safety for body weight of 10 mg of supplemental dapagliflozin in combination with metformin (24-week confirmatory study + 78-week long-term extension period).

-4.54 ± 0.4499 kg ($n = 69$) in the dapagliflozin 10 mg group at Week 102. Weight decreased associated with dapagliflozin was increased until Week 50, and remained until completion of the study (Week 102). The change in fat mass from baseline to Week 102 (adjusted mean \pm SE) was -1.46 ± 0.3985 kg ($n = 71$) in the placebo group and -2.80 ± 0.4403 kg ($n = 66$) in the 10 mg group. The change in lean mass from baseline to Week 102 (adjusted mean \pm SE) was -0.9 ± 0.226 kg ($n = 71$) in the placebo group and -1.3 ± 0.253 kg ($n = 67$) in the dapagliflozin 10 mg group. Therefore, loss of fat mass and loss of lean mass (including volume depletion) were estimated to account for two-thirds and one-third, respectively, of the weight decreased.

An evaluation of mean change from baseline in body weight revealed that the body weight in the dapagliflozin groups in Japanese Study D1692C00006 was substantially decreased after 1 week of treatment, followed by a gradual decrease until 16 weeks of treatment and little change thereafter; the difference [two-sided 95% CI] in the adjusted mean change from baseline to Week 24 of treatment was $-1.29 [-1.98, -0.59]$ kg between the placebo and the dapagliflozin 5 mg group and $-1.38 [-2.08, -0.69]$ kg between the placebo and the dapagliflozin 10 mg groups. In Japanese Study D1692C00012, the mean change from baseline peaked (-2.61 kg) at Week 52 in the monotherapy group. The mean change from baseline peaked (-1.88 kg) at Week 52 in the dapagliflozin + SU group, peaked (-2.61 kg) at Weeks 32 and 52 in the dapagliflozin + DPP-4 group, peaked (-2.79 kg) at Week 52 in the dapagliflozin + α -GI group, and peaked (-2.49 kg) at Week 52 in the dapagliflozin + BG group. In the dapagliflozin + TZD group, body weight decreased by 0.64 to 1.48 kg on average throughout the treatment period, and the change from baseline to Week 52 of treatment was -0.64 kg, with the peak (-1.48 kg) at Week 12. The mean change from baseline peaked (-2.50 kg) at Week 52 in the dapagliflozin + glinide group, and peaked (-3.01 kg) at Week 52 in the dapagliflozin + GLP-1 group. The change (mean \pm SD) in body weight by BMI (kg/m²) from baseline to Week 52 of treatment in the monotherapy group in Japanese Study D1692C00012 was -0.93 ± 1.827 ($n = 7$) in subjects with BMI <20 kg/m², -2.15 ± 2.009 ($n = 115$) in subjects with BMI of ≥ 20 and <25 kg/m², -2.70 ± 2.183 ($n = 88$) in subjects with BMI of ≥ 25 and <30 kg/m², and -3.88 ± 2.800 ($n = 39$) in subjects with BMI ≥ 30 kg/m². The incidence of adverse events by BMD during 52 weeks of treatment in the monotherapy group in Japanese Study D1692C00012 was 85.7% (6 of 7 subjects) with BMI <20 kg/m², 74.8% (86 of 115 subjects) with BMI of ≥ 20 and <25 kg/m², 85.2% (75 of 88 subjects) with BMI of ≥ 25 and <30 kg/m², and 76.9% (30 of 39 subjects) with BMI ≥ 30 kg/m². The incidence of adverse events related to volume depletion was 0% (0 of 7 subjects) with BMI <20 kg/m², 0.9% (1 of 115 subjects) with BMI of ≥ 20 and <25 kg/m², 2.3% (2 of 88 subjects) with BMI of ≥ 25 and <30 kg/m², and 0% (0 of 39 subjects) with BMI ≥ 30 kg/m². Although the number of subjects with BMI <20 kg/m² was generally limited, there was no trend toward a higher incidence of adverse events in patients with lower BMI.

Since weight decreased associated with dapagliflozin has been observed and the number of studied subjects with low BMI was limited, PMDA considers that it is necessary to continue to collect information on weight decreased via post-marketing surveillance.

4.(iii).B.(3).7 Renal disorder

The applicant explained as follows:

In the main Japanese and foreign clinical studies, treatment discontinuation criteria for high serum creatinine values were established in order to discontinue study treatment in patients who reported abnormality of renal function tests or renal failure specified by the criteria by considering the decreased renal function as an adverse event. In Japanese Study D1692C00005, renal disorder/renal failure-related events were not reported. In Japanese Study D1692C00006, renal disorder/renal failure-related events were reported by 3 subjects in the placebo group, 2 subjects in the dapagliflozin 5 mg group, and 6 subjects in the 10 mg group, but no events were serious. The studies included patients with eGFR (mL/min/1.73 m²) ≥ 45 at study entry, and treatment was to be discontinued in patients who reported eGFR <45 after the start of treatment by considering

the decrease as an adverse event. All subjects who discontinued treatment with dapagliflozin had moderate renal impairment with eGFR <60 at study entry and at baseline, and discontinued treatment due to a decrease in eGFR, but these values reversed or tended to reverse after treatment discontinuation. In Study D1692C00012, the incidence of renal disorder/renal failure-related events during 52 weeks of treatment was 2.0% (5 of 249 subjects) in the monotherapy group; 0.8% (1 of 122 subjects) in the dapagliflozin + SU group; 14.1% (10 of 71 subjects) in the dapagliflozin + BG group; 3.1% (2 of 64 subjects) in the dapagliflozin + TZD group; 2.0% (1 of 49 subjects) in the dapagliflozin + glinide group; and 4.0% (2 of 50 subjects) in the dapagliflozin + GLP-1 group. No such events were reported in the dapagliflozin + DPP-4 or α -GI groups. The renal disorder/renal failure-related adverse events included renal impairment (16 subjects), glomerular filtration rate decreased (4 subjects), and renal failure (1 subject); all except the 2 events of renal impairment (1 subject in the dapagliflozin + BG group, 1 subject in the dapagliflozin + glinide group) were reported because of meeting the treatment discontinuation criteria for eGFR. The incidence of renal disorder/renal failure-related adverse events was higher in the dapagliflozin + BG group, but this can be attributed to the range of eGFR used to identify adverse events for subjects in the dapagliflozin + BG group (<60), that was different from those for the other concomitant therapy groups (<45). Serious renal disorder/renal failure-related events were not reported.

In concomitant use with insulin, 1 event of nephrolithiasis was reported by 1 subject in the placebo group and 1 event of renal failure was reported by 1 subject in the dapagliflozin 10 mg group in Foreign Study MB102009.¹⁵⁰ The event of renal failure was severe and assessed as an adverse drug reaction, and led to treatment discontinuation [see “4.(iii).B.(3).5) Volume depletion” for details of this subject]. In Foreign Study D1690C00006,¹⁵¹ the incidence of renal disorder/renal failure-related events during 104 weeks of treatment was 2.0% (4 events in 4 of 197 subjects) in the placebo group; 1.5% (3 events in 3 of 202 subjects) in the dapagliflozin 2.5 mg group; 2.8% (9 events in 6 of 212 subjects) in the 5/10 mg group; and 3.1% (8 events in 6 of 196 subjects) in the 10 mg group, showing higher incidences in the dapagliflozin 5/10 mg and 10 mg groups. Severe events included renal failure acute (1 subject in the placebo group) and blood creatinine increased (1 subject in the dapagliflozin 5/10 mg group, 2 subjects in the 10 mg group). Events leading to treatment discontinuation included renal failure acute (1 subject in the placebo group), blood creatinine increased (1 subject in the dapagliflozin 2.5 mg group, 2 subjects in the 5/10 mg group, 3 subjects in the 10 mg group), and renal impairment (1 subject in the 5/10 mg group). Of 7 subjects who experienced an adverse event of blood creatinine increased, 5 subjects had a history of diabetic nephropathy, and 4 subjects had serum creatinine abnormally high.

Updated data from a pooled analysis (30-MU¹⁵³) showed that the incidence of renal disorder/renal failure-related events in 30-MU (short-term) was 1.8% (42 of 2295 subjects) in the placebo group and 3.2% (76 of 2360 subjects) in the dapagliflozin 10 mg group, showing a higher incidence in the dapagliflozin group [Table 26]. The main events included creatinine renal clearance decreased and renal impairment. Serious adverse events were reported by 1 subject in the placebo group (renal failure acute) and 2 subjects in the dapagliflozin 10 mg group (renal failure and renal failure acute in 1 subject each). In both the placebo and dapagliflozin groups, subjects aged ≥ 65 years accounted for over half of subjects (27 of 42 subjects [64.3%] in the placebo group, 51 of 76 subjects [67.1%] in the dapagliflozin 10 mg group) who experienced renal disorder/renal failure-related events, and subjects with moderate renal impairment (eGFR of ≥ 30 and <60) accounted for approximately 60% of subjects (25 of 42 subjects [59.5%] in the placebo group, 49 of 76 subjects [64.5%] in the dapagliflozin 10 mg group) who experienced renal disorder/renal failure-related events. The incidences of renal disorder/renal failure-related events by baseline eGFR were 9.3% (25 of 268 subjects) in the placebo group and 18.5% (49 of 265 subjects) in the dapagliflozin 10 mg group in subjects with baseline eGFR of ≥ 30 and <60; and 0.8% (17 of 2025 subjects) in the placebo group and 1.3% (27 of 2094 subjects) in the dapagliflozin 10 mg group in subjects with baseline eGFR ≥ 60 , showing a higher incidence in patients with moderate renal

impairment and a higher incidence in the dapagliflozin group than in the placebo group. In both groups, approximately 20% of subjects who experienced renal disorder/renal failure-related events showed a decreased eGFR. In both groups, the majority (>80%) of subjects who experienced renal disorder/renal failure-related events showed an eGFR of ≥ 0.85 -fold the baseline value during treatment or follow-up period after the event. The proportion of subjects who discontinued treatment due to renal disorder/renal failure-related adverse events was 1.2% (27 subjects) in the placebo group and 1.9% (44 subjects) in the dapagliflozin 10 mg group in the pooled population from short-term placebo-controlled studies; of these, the proportion of subjects in whom the event persisted was approximately 25% in both groups (25.9% [7 of the 27 subjects] in the placebo group, 25.0% [11 of the 44 subjects] in the dapagliflozin 10 mg group). There was no consistent trend in the time to onset or outcome of adverse events leading to treatment discontinuation.

The incidence of renal disorder/renal failure-related events in 30-MU (short-term + long-term) was 4.2% (82 of 1956 subjects) in the placebo group and 6.7% (136 of 2026 subjects) in the dapagliflozin 10 mg group, showing a higher incidence in the dapagliflozin group [Table 26]. The proportion of subjects who discontinued treatment due to renal disorder/renal failure-related adverse events was 2.8% (54 subjects) in the placebo group and 4.1% (83 subjects) in the dapagliflozin 10 mg group; of these, the proportion of subjects in whom the event persisted was 48.1% (26 of the 54 subjects) in the placebo group and 36.1% (30 of the 83 subjects) in the dapagliflozin 10 mg group. There was no consistent trend in the time to onset or outcome of adverse events leading to treatment discontinuation.

Table 26. Incidence of adverse events of renal disorder or renal failure in 30-MU (pooled population from short-term placebo-controlled studies)

Event term	30-MU (short-term)		30-MU (short-term + long-term)	
	Placebo group (n = 2295)	Dapagliflozin 10 mg group (n = 2360)	Placebo group (n = 1956)	Dapagliflozin 10 mg group (n = 2026)
Renal disorder or renal failure	42 (1.8)	76 (3.2)	82 (4.2)	136 (6.7)
Creatinine renal clearance decreased	16 (0.7)	27 (1.1)	28 (1.4)	46 (2.3)
Renal impairment	12 (0.5)	20 (0.8)	21 (1.1)	39 (1.9)
Blood creatinine increased	9 (0.4)	15 (0.6)	16 (0.8)	24 (1.2)
Glomerular filtration rate decreased	3 (0.1)	7 (0.3)	8 (0.4)	11 (0.5)
Renal failure	2 (0.1)	4 (0.2)	7 (0.4)	11 (0.5)
Renal failure acute	1 (0.0 ^a)	3 (0.1)	2 (0.1)	4 (0.2)
Cystatin C increased	0 (0.0)	2 (0.1)	0 (0.0)	3 (0.1)
Acute prerenal failure	0 (0.0)	1 (0.0 ^a)	0 (0.0)	0 (0.0)
Creatinine renal clearance abnormal	1 (0.0 ^a)	1 (0.0 ^a)	1 (0.1)	3 (0.1)
Renal function test abnormal	0 (0.0)	1 (0.0 ^a)	0 (0.0)	1 (0.0)
Urine flow decreased	0 (0.0)	1 (0.0 ^a)	0 (0.0)	3 (0.1)
Urine output decreased	1 (0.0 ^a)	0 (0.0)	1 (0.1)	1 (0.0 ^a)
Glomerular filtration rate abnormal	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.0 ^a)
Renal failure chronic	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.0 ^a)
Anuria	0 (0.0)	0 (0.0)	1 (0.1)	0 (0.0)

Number of subjects with even (incidence %), MedDRA/J ver.15.1

a) One subject experienced this event, but the value is reported as 0.0 after rounding to 1 significant digit.

Regarding effects on laboratory parameters, no notable laboratory abnormalities related to renal function were reported by any subject in Japanese Study D1692C00005. In Japanese Study D1692C00006, there were no subjects with BUN of ≥ 60 mg/dL or urea of >21.4 mmol/L. In addition, there were no subjects with serum creatinine of ≥ 1.5 -fold the baseline value or of ≥ 2.5 mg/dL. In Japanese Study D1692C00012, 1 subject in the Dapagliflozin + SU group showed an increased serum creatinine level of ≥ 1.5 -fold the baseline value. There were no subjects with BUN ≥ 60 mg/dL, urea >21.4 mmol/L, or serum creatinine ≥ 2.5 mg/dL. Notable laboratory

abnormalities (urine albumin/creatinine ratio of >1800 mg/g) were reported by 1 subject each in the monotherapy group, dapagliflozin + SU, and dapagliflozin + GLP-1 groups.

In 30-MU (short-term), no changes were observed in the mean change from baseline in eGFR, BUN, serum creatinine, or haematocrit in the placebo group throughout the 24-week treatment period. Change from baseline (mean \pm SE) in eGFR in the dapagliflozin 10 mg group decreased at Week 1 (-4.2 ± 0.30 mL/min/1.73m² [n = 1102]), followed by a gradual increase thereafter (-1.4 ± 0.27 mL/min/1.73m² [n = 1954] at Week 24). For BUN, the change in the dapagliflozin 10 mg group increased at Week 1 (1.7 ± 0.095 mg/dL [n = 1998]), followed by a similar level of increase thereafter until Week 24 (1.5 ± 0.097 mg/dL [n = 1956]). For serum creatinine, the change in the dapagliflozin 10 mg group increased at Week 1 (0.041 ± 0.0029 mg/dL [n = 1112]), followed by a decrease thereafter (0.019 ± 0.0026 mg/dL [n = 1954] at Week 24). For haematocrit, the change in the dapagliflozin 10 mg group continued to increase from Week 1 ($0.55\% \pm 0.045\%$ [n = 1989]) to Week 16 ($2.32\% \pm 0.056\%$ [n = 1932]), and was maintained from Week 16 on. The mean change from baseline (mean \pm SD) in systolic blood pressure, in the placebo group, slightly decreased at Week 1 (-0.9 ± 10.83 mmHg [n = 1888]) and was maintained until Week 12, followed by a gradual increase to near baseline values thereafter (-0.5 ± 13.08 mmHg [n = 1880] at Week 24). In the dapagliflozin 10 mg group, the change decreased at Week 1 (-3.3 ± 11.72 mmHg [n = 1940]), followed by a similar level of decrease thereafter until Week 24 (-3.7 ± 13.64 mmHg [n = 1987]). Diastolic blood pressures showed decreases similar to those in systolic blood pressure; diastolic blood pressure in the placebo group also had a slight decrease at Week 1 (-0.5 ± 7.04 mmHg [n = 1888]), followed by a similar level of decrease thereafter until Week 24 (-0.5 ± 8.47 mmHg [n = 1880]), while the value in the dapagliflozin 10 mg group decreased at Week 1 (-1.5 ± 7.17 mmHg [n = 1940]), followed by a similar level of decrease thereafter until Week 24 (-1.8 ± 8.01 mmHg [n = 1987]). Little change in the mean change from baseline in body weight was observed in the placebo group, but the value decreased from Week 1 (was -0.8 ± 0.03 kg [n = 1638]) until Week 24 (-2.3 ± 0.07 kg [n = 1815]) throughout the treatment period in the dapagliflozin 10 mg group.

As described above, neither eGFR changed throughout the 24-week treatment period nor clinically meaningful changes over time were seen in BUN, haematocrit, or body weight in the placebo group, while in the dapagliflozin group, a decrease in eGFR and increases in BUN and haematocrit as well as decreases in body weight and blood pressure were observed at 1 week after the start of treatment. Thus, the decrease in eGFR observed in the dapagliflozin group at an early stage of treatment was likely to be caused by hemodynamic regulation associated with diuresis, tubuloglomerular feedback, and decreased blood pressure. Then, eGFR tended to reverse during the treatment period, but BUN and blood pressure continued to change with a similar trend throughout the 24-week period, and a further increase was observed in haematocrit. Given the trend toward reversibility of eGFR observed during the treatment period, dapagliflozin is unlikely to exacerbate renal impairment. Therefore, the risk of renal disorder or renal failure due to dapagliflozin is considered to be limited. However, a careful follow-up/monitoring regarding renal disorder and renal failure will be continued in the future considering the mechanism of action of dapagliflozin.

PMDA considers as follows:

Given the observed findings such as increases in BUN and haematocrit and a decrease in blood pressure, the decrease in eGFR observed at an early stage of treatment with dapagliflozin is likely to be due to the effect of volume depletion. The incidence of renal disorder/renal failure-related events tended to be higher in patients with moderate renal impairment. However, long-term effects of continued treatment with dapagliflozin in patients with decreased eGFR <45 have not been adequately evaluated because treatment was planned to be discontinued in subjects in whom eGFR was decreased to <45 in the main Japanese and foreign clinical studies. Based on the above, it is necessary to advise that regular renal function tests be performed and to continue to collect

information on renal disorder via post-marketing surveillance [see “4.(iii).B.(6).1) Patients with renal impairment” for safety in patients with renal impairment].

4.(iii).B.(3).8 Increase in ketone bodies

The applicant explained as follows:

In Foreign Study MB102001 evaluating single administration of dapagliflozin at doses up to 500 mg and Foreign Study MB102002 evaluating once daily administration of dapagliflozin at doses up to 100 mg for 2 weeks, urine ketone bodies were measured at baseline and major timepoints after the start of treatment. As a result, almost no urine ketone bodies were detected and no signs of ketosis were observed. Based on the above, only 2 of the subsequent clinical studies (a foreign phase II study [Study MB102045¹⁶³], a foreign phase III study [Study MB102032¹⁶⁴]) of dapagliflozin included measurement of urine ketone bodies. In Study MB102045, the change (mean ± SD) from baseline to Week 12 of treatment in fasting serum acetoacetate was $-0.9 \pm 3.07 \mu\text{g/mL}$ in the placebo group and $-1.1 \pm 4.28 \mu\text{g/mL}$ in the dapagliflozin 5 mg group. The change (mean ± SD) in serum β -hydroxybutyric acid was $-3.8 \pm 9.36 \mu\text{g/mL}$ in the placebo group and $0.8 \pm 9.64 \mu\text{g/mL}$ in the dapagliflozin 5 mg group. In Study MB102032, the adjusted mean change (LOCF) from baseline to Week 24 of treatment in fasting acetoacetate was $0.20 \mu\text{g/mL}$ in the placebo group and $0.47 \mu\text{g/mL}$ in the dapagliflozin group, showing a greater increase in the dapagliflozin group. The difference [two-sided 95% CI] from placebo in the adjusted mean change from baseline to Week 24 of treatment in β -hydroxybutyric acid was $0.53 [-6.21, 7.28] \mu\text{g/mL}$ in the dapagliflozin 1 mg group, $1.05 [-5.77, 7.87] \mu\text{g/mL}$ in the 2.5 mg group, and $5.29 [-1.66, 12.25] \mu\text{g/mL}$ in the 5 mg group. No events of ketoacidosis or hyperglycaemia were reported in the dapagliflozin groups in both studies.

PMDA asked the applicant to explain the possibility that dapagliflozin may induce acute diabetic complications such as ketoacidosis due to its mechanism of action.

The applicant responded as follows:

Ketoacidosis is caused by ketone bodies produced by enhanced lipolysis in starved cells with inadequate glucose uptake via insulin action. Evaluation of ketone bodies can be performed by measuring β -hydroxybutyric acid, a by-product of lipolysis. Although β -hydroxybutyric acid is a substance commonly detected along with weight decreased and ketoacidosis, the two conditions have different mechanisms and clinical relevance. Diet therapy such as energy restriction or an insulin deficient state results in fat combustion to get an energy source and in production of ketone bodies. Ketonuria is a frequently observed finding in those with weight decreased, and in some cases, ketonuria can be used to predict weight decreased caused by the energy-restricted diet (Kim HJ, et al. *J Korean Med Sci*. 2012;27:250-4). A more than 6-fold increase in β -hydroxybutyric acid has been reported to be observed in nondiabetic obese subjects on the very low energy diet (500-550 kcal/day) for 8 weeks (Sumithran P, et al. *Eur J Clin Nutr*. 2013;67:759-64). However, β -hydroxybutyric acid level observed in the dapagliflozin 5 mg group at Week 24 in Foreign Study MB102032 was $19.10 \mu\text{g/mL}$, which was 1.3-fold the baseline level ($14.21 \mu\text{g/mL}$). This slight increase in ketone body level was below the upper limit of normal (approximately 4 mg/dL), and far below the level requiring intervention (10 mg/dL) (Wallace TM, et al. *Diabet Med*. 2001;18:640-5).

In Foreign Study MB102008,¹²⁷ following multiple doses of dapagliflozin 5 or 10 mg for 12 weeks in foreign patients with type 2 diabetes mellitus, approximately 70 g of glucose was

¹⁶³ Foreign Study MB102045: A placebo-controlled, randomized, double-blind, parallel-group phase IIb study that evaluated the effects of 5 mg of supplemental dapagliflozin on insulin sensitivity in patients with type 2 diabetes mellitus who received treatment with metformin and/or an insulin secretagogue (treatment duration was 12 weeks).

¹⁶⁴ Foreign Study MB102032: A placebo-controlled, randomized, double-blind, parallel-group phase III study that evaluated the efficacy and safety of dapagliflozin monotherapy at doses of 1, 2.5, and 5 mg in patients with type 2 diabetes mellitus without a history of antidiabetic medication (treatment duration was 24 weeks).

excreted in urine, which is equivalent to an energy loss of approximately 10% (280 kcal) of the total daily energy intake in adults. In general, patients with diabetes mellitus obtain energy equivalent to 50% to 60% of the total daily energy intake in the form of carbohydrates (The Japan Diabetes Society, ed. *Treatment Guide for Diabetes* 2012-2013). Therefore, dapagliflozin is unlikely to induce a shortage of carbohydrates because an energy loss of approximately 10% possibly caused by dapagliflozin would not be substantial, and glucose remains as the main energy source.

Updated data from a pooled analysis (30-MU¹⁵³) showed that only 1 event of ketoacidosis was reported by 1 subject in the dapagliflozin group (Study D1690C0006¹⁵¹ evaluating combination therapy with insulin) in 30-MU (all studies) (3403 subjects in the placebo group, 5936 subjects in the dapagliflozin group). This subject was a 52-year-old white female complicated with hypertension and dyslipidaemia receiving metformin and insulin therapy, and hospitalized due to gastroenteritis on Day 213 and vomiting, diarrhoea, dehydration, acidosis, weakness, and discomfort on Day 214. The subject received a diagnosis of gastroenteritis and diabetic ketoacidosis, and additionally on Day 214, of asymptomatic bacteriuria. Study treatment was temporarily discontinued from Day 215 to Day 217, but resumed after recovery on Day 218. In addition, in the same pooled population, 2 events of ketonuria were reported by 2 subjects in the dapagliflozin 10 mg group, but the events were non-serious and treatment was continued in both subjects.

As described in the above, only 1 event of ketoacidosis was reported across all clinical studies and a causal relationship to the study drug was ruled out for the event, and no signs leading to ketoacidosis were observed, thus, the risk of dapagliflozin-related ketoacidosis is limited.

PMDA accepted the applicant's response. However, given the concern that dapagliflozin may induce acute diabetic complications associated with an increase in ketone bodies in addition to dapagliflozin-related volume depletion in patients with type 2 diabetes mellitus with reduced insulin secretion [see "4.(iii).B.(3).5) Volume depletion" for volume depletion], it is necessary to continue to collect information on increase in ketone bodies via post-marketing surveillance.

4.(iii).B.(3).9 Impact on bone metabolism

The applicant explained as follows:

Because dapagliflozin may have an impact on body weight and absorption/excretion of Ca and P in the renal tubules through its pharmacological activity, bone mineral density was evaluated in Foreign Study D1690C00012¹⁶² by bone mineral determination using dual-energy X-ray absorptiometry (DXA). The results showed no meaningful changes in the percent change from baseline to Week 102 of treatment in both the dapagliflozin and placebo groups. In addition, analyses by sex and eGFR revealed that no meaningful changes in bone mineral density were observed in any group. Decreases in the mean levels of all markers for bone formation (osteocalcin [OC], bone-specific alkaline phosphatase [BAP], N-terminal propeptide of type 1 procollagen [P1NP]) were observed in the placebo group, but no meaningful changes were seen from baseline to Week 102 of treatment in the dapagliflozin groups. Little changes were observed in markers for bone resorption (type 1 collagen-crosslinked C-telopeptide [CTX], type 1 collagen-crosslinked N-telopeptide [NTX]).

In Japanese Study D1692C00006, fractures were reported by 1 subject in the placebo group (wrist fracture), 1 subject in the dapagliflozin 5 mg group (foot fracture), and 2 subjects in the 10 mg group (hand fracture, rib fracture), but a causal relationship to the study drug was ruled out for all these events. Rib fracture reported in the dapagliflozin 10 mg group was assessed as a serious adverse event. Dapagliflozin did not induce meaningful changes in parathyroid hormone, serum Ca, or serum inorganic P. Slightly greater increases from baseline in serum Mg were observed in the dapagliflozin groups (0.09 mEq/L in both 5 and 10 mg groups) than in the placebo group (0.02

mEq/L), but these values returned to baseline during the follow-up period. In Japanese Study D1692C00012, the incidence of fracture was 2.4% (6 of 249 subjects) in the monotherapy group; 2.5% (3 of 122 subjects) in the dapagliflozin + SU group; 1.6% (1 of 61 subjects) in the dapagliflozin + α -GI group; 1.6% (1 of 64 subjects) in the dapagliflozin + TZD group; and 2.0% (1 of 50 subjects) in the dapagliflozin + GLP-1 group. Fracture was not reported in the dapagliflozin + DPP-4, BG, or glinide groups. Notable laboratory abnormalities involving serum electrolyte were rarely reported. No important changes were observed in serum Ca or serum inorganic P throughout the treatment period in any group. A slight increase from baseline in mean serum Mg levels was observed after 52 weeks of treatment (0.05 mEq/L in the monotherapy group), but this value returned to baseline during the follow-up period. A slight increase from baseline in mean parathyroid hormone levels was observed after 52 weeks of treatment (0.8 pg/mL in the monotherapy group), but this value was reversed during the follow-up period.

In concomitant use with insulin, the incidence of fracture during 104 weeks of treatment in Foreign Study D1690C00006¹⁵¹ was 3.0% (6 events in 6 of 197 subjects) in the placebo group; 2.0% (4 events in 4 of 202 subjects) in the dapagliflozin 2.5 mg group; 3.3% (7 events in 7 of 212 subjects) in the 5/10 mg group; and 3.6% (8 events in 7 of 196 subjects) in the 10 mg group. A causal relationship to the study drug was ruled out for all these events.

Updated data from a pooled analysis (30-MU¹⁵³) showed that the incidence of fracture in 30-MU (short-term) was 0.7% (17 of 2295 subjects) in the placebo group and 0.3% (8 of 2360 subjects) in the dapagliflozin 10 mg group; and the incidence of fracture in 30-MU (short-term + long-term) was 1.6% (32 of 1956 subjects) in the placebo group and 1.1% (23 of 2026 subjects) in the dapagliflozin 10 mg group, showing a low incidence in both pooled populations and lower incidences in the dapagliflozin groups than in the placebo group. No relationship was observed between dapagliflozin and clinically significant changes in markers for bone formation and resorption. In 30-MU (short-term), no clinically meaningful changes in mean serum P levels were observed after 24 weeks of treatment in the placebo (-0.04 mg/dL) or in dapagliflozin 10 mg (0.13 mg/dL) groups. No clinically meaningful changes in mean serum Ca levels were observed after 24 weeks of treatment in the placebo (-0.01 mg/dL) or in dapagliflozin 10 mg (0.04 mg/dL) groups. The mean level of serum Mg after 24 weeks of treatment was slightly higher in the dapagliflozin 10 mg group (0.09 mEq/L) than in the placebo group (-0.02 mEq/L). The mean level of parathyroid hormone after 24 weeks of treatment was slightly higher in the dapagliflozin 10 mg group (4.065 pg/mL) than in the placebo group (1.358 pg/mL). No clinically meaningful changes in mean 25-hydroxyvitamin D levels were observed after 24 weeks of treatment in the placebo (-1.144 ng/mL) or in dapagliflozin 10 mg (-0.137 ng/mL) groups.

Since the number of subjects and treatment duration studied in clinical trials were limited, PMDA considers that it is necessary to continue to collect information on the impact on bone metabolism via post-marketing surveillance.

4.(iii).B.(3).10 Cardiovascular risk

The applicant explained as follows:

Adverse events classified as System Organ Class “cardiac disorders” or “vascular disorders” in Japanese Study D1692C00006 include hypertension (5 subjects in the placebo group, 2 subjects in the dapagliflozin 5 mg group, 1 subject in the 10 mg group), blood pressure inadequately controlled (1 subject in the 5 mg group), and atrial fibrillation (1 subject in the 10 mg group). In Japanese Study D1692C00012, the incidence of adverse events classified as “cardiac disorders” was 2.4% (6 of 249 subjects) in the monotherapy group; 0.8% (1 of 122 subjects) in the dapagliflozin + SU group; 4.8% (3 of 62 subjects) in the dapagliflozin + DPP-4 group; 1.6% (1 of 61 subjects) in the dapagliflozin + α -GI group; 1.6% (1 of 64 subjects) in the dapagliflozin + TZD group; and 4.1% (2 of 49 subjects) in the dapagliflozin + glinide group. No such events were reported in the dapagliflozin + BG and GLP-1 groups. The events reported by 3 subjects in the

monotherapy group (arrhythmia/ventricular extrasystoles, supraventricular extrasystoles, tachycardia paroxysmal), 1 subject in the dapagliflozin + SU group (hypertrophic cardiomyopathy), 1 subject in the dapagliflozin + α -GI group (bundle branch block right), and 1 subject in the dapagliflozin + glinide group (arrhythmia) were assessed as adverse drug reactions. The incidence of adverse events classified as “vascular disorders” was 2.0% (5 of 249 subjects) in the monotherapy group; 4.8% (3 of 62 subjects) in the dapagliflozin + DPP-4 group; 1.6% (1 of 61 subjects) in the dapagliflozin + α -GI group; 2.8% (2 of 71 subjects) in the dapagliflozin + BG group; 2.0% (1 of 49 subjects) in the dapagliflozin + glinide group; and 2.0% (1 of 50 subjects) in the dapagliflozin + GLP-1 groups. No such events were reported in the dapagliflozin + SU and + TZD groups. The events reported by 2 subjects in the monotherapy group (hypertension, orthostatic hypotension) were assessed as adverse drug reactions.

No clinically meaningful changes were observed in the mean change in heart rate from baseline to Week 24 of treatment in any group in Study D1692C00006. Neither clinically meaningful changes in the mean change in heart rate from baseline to Week 52 of treatment nor increasing trend in the degree of changes with increasing treatment duration were observed in any group in Study D1692C00012.

Regarding blood pressure, in Study D1692C00006, a slight decrease in the mean change from baseline to Week 24 of treatment in systolic blood pressure was observed in the dapagliflozin group, which reversed during the follow-up period. No clinically meaningful changes were observed in diastolic blood pressure. In Study D1692C00012, a slight decrease in the mean change from baseline to Week 52 of treatment in systolic blood pressure was observed in all groups, which reversed during the follow-up period. Systolic blood pressure showed the lowest value in the dapagliflozin + GLP-1 group, with the mean change from baseline of -3.6 to 0.6 mmHg. No clinically meaningful changes were observed in the mean change from baseline for diastolic blood pressure.

Regarding lipids, in Japanese Study D1692C00006, no clinically meaningful changes were observed in the change from baseline in the mean fasting lipids. In the monotherapy group in Japanese Study D1692C00012, the mean percent changes from baseline to Week 52 of treatment in total cholesterol (T-chol), LDL cholesterol (LDL-C), HDL cholesterol (HDL-C), and free fatty acid (FFA) were 3.7%, 4.4%, 9.7%, and 6.0%, respectively, while that in triglycerides (TGs) was -8.8%. Similarly, the mean percent changes from baseline to Week 52 of treatment in T-chol, LDL-C, HDL-C, and FFA were 4.1%, 3.9%, 8.6%, and 14.2%, respectively, while that in TGs was -5.6% in the dapagliflozin + SU group; 5.9%, 6.8%, 10.2%, and 10.9%, respectively, and that in TGs of -8.3% in the dapagliflozin + DPP-4 group; 1.8%, 0.7%, 9.1%, and 5.8%, respectively, and that in TGs of -8.9% in the dapagliflozin + BG group; 3.4%, 3.0%, 7.2%, and 11.8%, respectively, and that in TGs of -5.6% in the dapagliflozin + TZD group; 3.3%, 5.3%, 7.3%, and 7.5%, respectively, and that in TGs of -12.1% in the dapagliflozin + glinide group; and 2.9%, 2.0%, 9.9%, and 16.1%, and that in TGs of -9.4% in the dapagliflozin + GLP-1 group. In the dapagliflozin + α -GI group, the percent changes at Week 52 in T-chol, LDL-C, and HDL-C, were 2.6%, 0.1%, and 9.6%, respectively, while that in TGs was -6.0%, and that in FFA increased and decreased repeatedly with a mean change of -1.3% to 11.3% throughout the treatment period, with the mean change at Week 52 of -1.3%.

In Study D1692C00006, no important changes were observed in ECG. In Study D1692C00012, 83.8% of subjects showed a normal ECG at baseline, and many subjects (91.0%) showed a normal ECG also at Week 52; an abnormal ECG was observed in 4.8% of subjects (3.7% [8 of 215 subjects] in the monotherapy group, 3.9% [4 of 103 subjects] in the dapagliflozin + SU group, 3.5% [2 of 57 subjects] in the dapagliflozin + DPP-4 group, 2.1% [1 of 48 subjects] in the dapagliflozin + α -GI group, 1.8% [1 of 56 subjects] in the dapagliflozin + BG group, 7.1% [4 of

56 subjects] in the dapagliflozin + TZD group, 15.2% [5 of 33 subjects] in the dapagliflozin + glinide group, and 9.5% [4 of 42 subjects] in the dapagliflozin + GLP-1 group).

Using updated data from a pooled analysis (30-MU¹⁵³), a meta-analysis¹⁶⁵ was performed of all cardiovascular events that were determined by blinded central review for 9339 subjects in 30-MU (all studies) (5936 subjects in all dapagliflozin groups, 3403 subjects in the control group). The estimated hazard ratio [two-sided 95% CI] for cardiovascular events (cardiovascular death, myocardial infarction, stroke, hospitalization due to unstable angina) in the dapagliflozin group was 0.787 [0.579, 1.070] compared with the control group, and that for major cardiovascular events (MACE; cardiovascular death, myocardial infarction, stroke) was 0.772 [0.543, 1.097].

In the combined population of the previously conducted 2 clinical studies of dapagliflozin in patients with type 2 diabetes mellitus who had a history of cardiovascular disease (Foreign Studies D1690C00018,¹⁶⁶ D1690C00019;¹⁶⁰ also included in the meta-analysis), the mean age was 63 years (subjects aged ≥65 years accounted for approximately 45%), mean BMI was 33 kg/m², and mean duration of diabetes mellitus was 13 years (subjects with ≥10 years of disease accounted for approximately 57%). At baseline, 84% of subjects had dyslipidaemia, 14% of subjects had cardiac failure congestive, and 96% of subjects had hypertension. In addition, almost all subjects had a history of cardiovascular disease other than hypertension. Subjects with eGFR of ≥60 and <90 accounted for approximately 60%, and subjects with eGFR of ≥30 and <60 accounted for 17% of the population. The meta-analysis revealed the estimated hazard ratio for cardiovascular events in subjects with a history of cardiovascular disease of 0.806 [0.562, 1.156], similar results to that obtained from the overall analysis.

Based on the above, at present, no trend toward increasing cardiovascular risk has been observed after administration of dapagliflozin. An ongoing global study (Study D1693C00001¹⁶⁷) will assess cardiovascular risk.

Given the findings of increased serum lipids, the trend toward a higher incidence of events related to volume depletion after administration of dapagliflozin, and increased haematocrit in subjects in the dapagliflozin group [see “4.(iii).B.(3).5) Volume depletion”], as well as the limited number of subjects and treatment duration in Japanese clinical studies, PMDA considers that it is necessary to continue to collect information on cardiovascular risk via post-marketing surveillance and to facilitate an evaluation of dapagliflozin-related cardiovascular risk also using data from the ongoing Study D1693C00001.

4.(iii).B.(3).11 Malignant tumor risk

The applicant explained as follows:

In Japanese Study D1692C00006, no developments of neoplasm were observed. In Japanese Study D1692C00012, the incidence of malignant and unspecified neoplasms¹⁶⁸ was 2.0% (5 of 249 subjects) in the monotherapy group and 1.6% (2 of 122 subjects) in the dapagliflozin + SU group; no such events were reported in the other concomitant therapy groups.

Updated data from a pooled analysis (30-MU¹⁵³) showed that the incidence per 100 person-

¹⁶⁵ Hazard ratio in each study included in the meta-analysis was estimated using the Cox proportional hazards model.

¹⁶⁶ Foreign Study D1690C00018: A placebo-controlled, randomized, double-blind, parallel-group phase III study in patients with type 2 diabetes mellitus on antihypertensive treatment for cardiovascular disease and hypertension that evaluated the efficacy and safety of 10 mg of supplemental dapagliflozin (consisting of a 24-week confirmatory study and the subsequent long-term extension periods [28-week + 52-week], for a total duration of 104 weeks).

¹⁶⁷ Global Study D1693C00001: An ongoing placebo-controlled, randomized, double-blind, parallel-group study in patients with type 2 diabetes mellitus at high cardiovascular risk (including Japanese patients) to evaluate the effects of dapagliflozin 10 mg on the incidences of cardiovascular death, myocardial infarction, and ischaemic stroke (target sample size, ≥17,000 including ■ Japanese subjects).

¹⁶⁸ Defined as events belonging to SMQ “Malignant and unspecified neoplasms.”

years [two-sided 95% CI] of malignant and unspecified neoplasms in 30-MU (all studies) was 1.35 [1.08, 1.66] in the dapagliflozin group and 1.33 [0.99, 1.75] in the control group, resulting in an incidence ratio [two-sided 95% CI] of 1.030 [0.711, 1.506] compared with the control group. Events with a relatively high incidence ratio (dapagliflozin group/control group) included bladder cancer and breast cancer.

The incidence of bladder cancer in 30-MU (all studies) combined with short-term treatment data from a clinical study that completed after the data cut-off date (Foreign Study D1693C00005) was 0.03% (1 of 3512 subjects) in the control group and 0.17% (10 of 6045 subjects) in the dapagliflozin group. The incidence per 100 person-years was 0.025 in the control group and 0.148 in the dapagliflozin group, resulting in an incidence ratio [two-sided 95% CI] of 6.111 [0.827, 272.02] compared with the control group. Except for 1 subject (a 53-year-old white female with a history of smoking 20 cigarettes per day for 40 years), all subjects who experienced bladder cancer were male. Of the 10 male subjects who experienced bladder cancer, 8 subjects (including 1 subject in the control group) were \geq 60 years of age, 8 subjects (including 1 subject in the control group) were a current smoker or had a history of smoking (of 2 subjects who were not a current or former smoker, 1 subject had a family history of bladder cancer); 8 subjects (including 1 subject in the control group) were white Europeans, and 2 subjects (including 1 Japanese subject) were Asians, showing similar characteristics as a whole, to that of the general population to have a diagnosis of bladder cancer. The Japanese subject who experienced bladder cancer (a 75-year-old male) had a history of smoking, and showed haematuria before and after the start of treatment; the time interval between the start of treatment and the onset of bladder cancer was 43 days. Nine of 10 subjects in the dapagliflozin group who experienced bladder cancer showed an evidence of haematuria at the start of the study treatment, and of these, a microscopic haematuria or a trace amount of haematuria was detected in 7 subjects within 6 months after the start of the study treatment. Based on the time of onset of haematuria in relation to the start of the study treatment, these subjects may have had bladder cancer before study entry. Haematuria was not observed in 1 subject from the start of treatment until \geq 6 months of treatment. All of the 11 subjects who experienced bladder cancer received study treatment for <2 years, and of these, 7 subjects (6 subjects in the dapagliflozin group, 1 subject in the control group) experienced bladder cancer within 1 year after the start of treatment (the onset time was <6 months in 6 subjects [including 1 subject in the control group], \geq 6 and <12 months in 1 subject, \geq 12 and <18 months in 2 subjects, \geq 18 and <24 months in 2 subjects). Of 11 subjects, 6 subjects could not be assessed by TNM staging (including 1 subject in the control group), 2 subjects were classified as pTa, 1 subject as T1, and 2 subjects as T2. Of 5 subjects who developed bladder cancer after \geq 6 months of treatment, 2 subjects could not be assessed by TNM staging, 2 subjects were classified as pTa, and 1 subject as T1. Dapagliflozin is unlikely to have newly induced a bladder cancer during the study period considering the long latency period (18-44 years) of bladder cancer induced by a certain chemical carcinogen (Mantanioski GM, et al. *Epidemiology Reviews*. 1981;3:203-29).

As described above, subjects in the dapagliflozin group in 30-MU who experienced bladder cancer may have experienced urinary tract-related symptoms due to diuretic effect of dapagliflozin, which led to the detection of the bladder cancer that may have preceded the treatment. In addition, no bladder tumors were observed in non-clinical studies, no mechanism can be assumed that explains the relationship with dapagliflozin, and the observation periods used in the clinical studies were limited, therefore, there is limited evidence to conclude that dapagliflozin affected the occurrence or proliferation of bladder cancer.

The incidence of breast cancer among female subjects in 30-MU (all studies) was 0.45% (12 of 2693 subjects) in the dapagliflozin group and 0.21% (3 of 1439 subjects) in the control group. The incidence per 100 person-years [two-sided 95% CI] of breast cancer was 0.19 [0.04, 0.56] in the control group and 0.40 [0.21, 0.70] in the dapagliflozin group, resulting in an incidence ratio

of 2.472 [0.636, 14.095] as compared with the control group. All subjects who experienced breast cancer in the dapagliflozin group were female aged >50 years (subjects aged ≥60 years accounted for 75% [9 of 12 subjects]) with ≥2 risk factors at baseline or a relevant history, and among these, postmenopausal subjects accounted for 92% (11 of 12 subjects) and overweight or obese subjects accounted for 83% (10 of 12 subjects), showing similar characteristics to those of the general population to have a diagnosis of breast cancer. The time interval between the start of treatment with dapagliflozin and diagnosis of breast cancer was <6 months in 4 subjects (including 2 subjects who received a diagnosis within 8 weeks) and 6 to <12 months in 9 subjects; thus, 13 of a total of 15 subjects received a diagnosis after <1 year of treatment. All events were reported after a short term exposure that was quite different from the reported latency period of human breast cancer induced by chemicals (several years to several decades) (Malone KE. *Epidemiologic Reviews*. 1993;15:108-9). Although a large variability exists, the mean doubling time of breast cancer has been reported to be approximately 150 days (Friberg S, et al. *J Surg Oncol*. 1997;65:284-97), which means that it takes 12 years for one malignant cell to grow to breast cancer of 1 cm in size. A very rapidly proliferating breast cancer doubles in size in 1 month and grows to 1 cm as early as 2.4 years later, but this period is considered longer than the time interval between the start of treatment with dapagliflozin and detection of cancer. Therefore, based on the estimations of cell doubling time and tumor size, the possibility that dapagliflozin may act as an initiator of breast cancer would be excluded. Generally, a rapidly proliferating breast cancer has morphological characteristics including a high dyskaryosis, a papillary growth pattern (Spratt JS, et al. *Cancer*. 1981;47:2265-8), a low incidence of oestrogen receptor positivity, a high incidence of axillary lymph node metastasis (Brekelmans CT, et al. *Cancer*. 1996;78:1220-8). None of these characteristics was clearly observed in any breast cancer reported in the dapagliflozin group, and in addition, no supporting data were presented in the non-clinical studies; therefore, the possibility that dapagliflozin act as a promoter would also be limited. The weight reducing effect of dapagliflozin may have promoted the detection of pre-existing cancerous lesions.

Based on the above, in spite of the difference in the incidences of bladder and breast cancers between the dapagliflozin and control groups observed in 30-MU, dapagliflozin is unlikely to have an impact on the difference, which is attributed to incidental detection of pre-existing tumors.

PMDA asked the applicant to explain the localization of SGLT isoforms in bladder epithelium and mammary gland tissues, the selectivity of dapagliflozin for each isoform, and the distribution of dapagliflozin in these tissues, and then explain the relationship between dapagliflozin and tumor risk.

The applicant responded as follows:

Based on an analysis on the expression of SGLT isoforms in a wide range of tissues performed by using a quantitative RT-PCR assay in a series of >70 normal human tissues including the bladder and mammary gland, only SMIT1 among SGLT isoforms has been reported to be expressed in bladder and mammary tissues at a substantial level; no expression of SGLT2 was observed in either tissue (Chen J, et al. *Diabetes Ther*. 2010;1:57-92). Dapagliflozin selectively inhibit SGLT2 compared with other human SGLTs. In a study using quantitative whole-body autoradiography, exposure to unchanged dapagliflozin or metabolites was seen in the bladder and mammary gland, but the levels of exposure were similar to those in other tissues or blood and no bladder- or mammary gland-specific retention of radioactivity was observed (4.2.2.3.3). An evaluation for cell proliferation promoting activity of dapagliflozin and dapagliflozin 3-O-glucuronide using 6 human bladder cancer cell lines showed no such activity (a preliminary study conducted by the applicant). In addition, tumor proliferation promoting activity was investigated by treating male and female nude mice (n = 8/dose) transplanted with a human bladder cancer cell line with dapagliflozin at doses of 0, 4, and 20 mg/kg (females) or at doses of 0, 12, and 60 mg/kg (males). The results showed no such activity (a preliminary study conducted by the applicant). In 2-year carcinogenicity studies, no carcinogenic potential was observed even at >46-

fold (male mice) or >100-fold (female rats) the exposure (AUC) at the maximum recommended clinical dose. Neither mutagenic potential nor *in vivo* clastogenic potential was observed. In the *in vitro* chromosomal aberration assay, no clastogenic potential was seen without metabolic activation, while clastogenic potential that would not jeopardize safety in humans was seen at concentrations of $\geq 100 \mu\text{g/mL}$ under metabolic activation [see “3.(iii).B.(3) Clastogenicity of metabolites”]. In a 15-month observation in SGLT2-KO mice, neither effects on the kidneys nor proliferative changes in the kidneys or bladder was observed in spite of the substantially high urinary glucose concentrations during the study period. Therefore, there is no evidence that dapagliflozin acts as a tumor initiator. Likewise, the potential for dapagliflozin to act as a tumor promoter was evaluated. In 2-year carcinogenicity studies in mice and rats, neither an increased incidence nor early onset of common tumors was observed after dapagliflozin treatment. For example, neither an increased incidence nor early onset of mammary gland tumors, which are commonly observed tumors in female rats, was observed after dapagliflozin treatment. Risk factors involved in stimulation of carcinogenesis primarily include immunosuppression, endocrine-disruption, inflammation, and stimulation of cell proliferation, and risk factors specifically related to bladder cancer may include changes in urine pH and constituents that may lead to crystalluria, irritability to the bladder, cytotoxicity, and cell proliferation. In the chronic toxicity studies of dapagliflozin in rats and dogs, no changes suggestive of a relationship with the above factors were found except for infrequent and mild inflammatory changes not leading to neoplastic changes. Generally, tumor promotion activity is caused by an enhanced cell proliferation. However, all toxicity studies including 2-year carcinogenicity studies in rodents (>46-fold [male mice] or >100-fold [female rats] the exposure [AUC] at the maximum recommended clinical dose) and 1-year repeated oral dose toxicity study in dogs (an animal species known as susceptible to bladder cancer) (>3000-fold the exposure [AUC] at the maximum recommended clinical dose) detected no hyperplasias or cell proliferative changes due to dapagliflozin in mammary gland, bladder, or other tissues. Therefore, there is no evidence that dapagliflozin acts as a tumor promoter.

Based on the above, the applicant considered the dapagliflozin-related risk of bladder and breast tumors as follows:

(a) There is no plausible biological mechanism that can explain the relationship between the occurrence of bladder or breast cancer and dapagliflozin; (b) Because of the limited observation periods used in the clinical studies, there is limited evidence to conclude that dapagliflozin affected the occurrence or proliferation of bladder or breast cancer; (c) Because bladder and breast cancers reported in the clinical studies reflected commonly observed clinical features, and especially, the majority of subjects who experienced bladder cancer showed haematuria before the start of treatment, the bladder cancer is likely to have already existed before the start of the study treatment; (d) The pharmacological activity of dapagliflozin may have expedited the detection of pre-existing cancerous lesions. In other words, the weight reducing effect of dapagliflozin may have promoted self-examination for breast cancer, or the urinary tract symptoms and urinary status that resulted from the diuretic effect of dapagliflozin may have expedited detection of pre-existing bladder cancer lesions.

A careful follow-up/monitoring of bladder and breast cancers will be conducted based on post-marketing data and data from Study D1693C00001,¹⁶⁷ etc.

PMDA considers as follows:

PMDA accepted the following applicant’s explanation (response): since these cancers were not observed in the non-clinical studies and dapagliflozin does not have the mechanism that can explain the relationship between the occurrence of bladder or breast cancer and dapagliflozin, there is little evidence to conclude that dapagliflozin affected the tumorigenesis based on the relationship between the observation period in the clinical studies and tumor growth rate. However, because it is difficult to conclude that the incidences of bladder and breast cancers were

accidentally higher than that in the control group in the clinical studies, it is necessary to continue to collect information on occurrence of tumors via post-marketing surveillance and to pay close attention to Japanese and foreign post-marketing and clinical study data, etc. The above issues will be finalized, taking account of comments from the Expert Discussion.

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

PMDA considers as follows:

“On release of the Guideline for Clinical Evaluation of Oral Hypoglycemic Agents” (PFSB/ELD Notification No. 0709-1 dated July 9, 2010, OAD Guideline) states that when an investigational drug is confirmed to be useful in clinical studies of 2-drug concomitant therapies with the investigational drug and the approved oral hypoglycemic agents (in combinations that are expected to be administered to patients in clinical practice) conducted based on OAD Guideline, the appropriate description of the indication is “type 2 diabetes mellitus.” Since the data submitted in this application demonstrate the efficacy of monotherapy and concomitant therapies through clinical studies conforming to OAD Guideline [see “4.(iii).B.(2) Efficacy”] and their safety is acceptable [see “4.(iii).B.(3) Safety”], PMDA considers that there is no problem with the proposed indication of “type 2 diabetes mellitus.”

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

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

The applicant explained that the frequency of administration of once daily is justified based on the plasma terminal half-life of dapagliflozin (12.9 hours) and the sustained inhibition of glucose reabsorption for 24 hours after the start of treatment.

Since morning dose regimens were evaluated in Japanese phase II and phase III studies (Studies D1692C00005, D1692C00006, and D1692C00012), PMDA asked the applicant to explain the impact of not specifying the timing of dosing (especially evening dosing) on the efficacy and safety.

The applicant responded as follows:

In a foreign phase III study (Study MB102013¹⁶⁹), a comparison was performed between morning and evening dose regimens [Table 27], and no substantial difference in the efficacy or safety of dapagliflozin was observed between the morning and evening groups. The incidence of nocturia was slightly higher in the dapagliflozin group (evening) than in the placebo group, but all events were mild in severity except for 1 moderate event reported in the 10 mg group. The timing of dosing was not found to be a significant covariate in a population pharmacokinetic analysis based on data from subjects in the morning and evening groups, and therefore was not included in the final model. The exposure range of dapagliflozin was similar between the morning and evening groups. In the food effect study (Study MB102062), no food effects were observed on AUC_{inf} of unchanged dapagliflozin, but the geometric mean of C_{max} of unchanged dapagliflozin after a high-fat meal decreased to up to approximately 55% of that under fasted conditions. AUC_{inf} was not affected in spite of the decrease in C_{max}, and the decrease in C_{max} is considered attributed to a decreased gastric emptying rate caused by a decreased absorption rate due to food intake, rather than to a change in total body clearance. C_{max} of unchanged dapagliflozin after fed administration of dapagliflozin 5 mg appeared to be similar to that after administration of dapagliflozin 2.5 mg under fasted conditions, and the urinary glucose excretion rate at around t_{max} after administration of dapagliflozin 2.5 mg under fasted conditions was not substantially different from that after administration of dapagliflozin 10 mg under fasted conditions (the urinary glucose excretion rate

¹⁶⁹ Study MB102013: A placebo-controlled, randomized, double-blind, parallel-group study in type 2 diabetes mellitus patients without a history of diabetes treatment that evaluated the efficacy and safety of dapagliflozin monotherapy (2.5, 5, or 10 mg) using a morning dose or evening dose regimen (24-week confirmatory study + 78-week long-term extension period).

between 0-4 hours post-dose on Day 14 in Study MB102002¹⁷⁰ was 0.89 ± 0.40 g/h in the 2.5 mg group and 1.25 ± 0.71 g/h in the 10 mg group). An evaluation of the relationship between the exposure (AUC) and change from baseline in 24-hour urinary glucose excretion revealed that this relationship fits a hyperbolic E_{max} model, as in the case of the relationship between the dose of dapagliflozin and change from baseline in 24-hour urinary glucose excretion. Therefore, given the facts that the urinary glucose excretion rate early after administration would not be affected by a decreased C_{max} after fed administration of dapagliflozin 5 mg, and that there was no food effect on AUC_{inf} , the impact of a decreased C_{max} due to food intake on the hypoglycemic activity is considered limited. Based on the above, the applicant considered that dapagliflozin may be administered at an unspecified time of day.

Table 27. Efficacy and safety during the double-blind period (24 weeks) of Foreign Study MB102013

	Placebo group (n = 75)	Dapagliflozin 2.5 mg group		Dapagliflozin 5 mg group		Dapagliflozin 10 mg group	
		Morning dosing (n = 65)	Evening dosing (n = 67)	Morning dosing (n = 64)	Evening dosing (n = 68)	Morning dosing (n = 70)	Evening dosing (n = 76)
Change in HbA1c (%)	-0.23 [-0.43, -0.02]	-0.58 [-0.80, -0.36]	-0.83 [-1.05, -0.61]	-0.77 [-0.99, -0.55]	-0.79 [-1.01, -0.57]	-0.89 [-1.10, -0.67]	-0.79 [-0.99, -0.59]
Between-group difference versus placebo	- [-0.65, -0.05]	-0.35 [-0.65, -0.30]	-0.61 [-0.91, -0.30]	-0.54 [-0.84, -0.24]	-0.56 [-0.86, -0.26]	-0.66 [-0.96, -0.36]	-0.56 [-0.85, -0.27]
All adverse events	45 (60.0)	41 (63.1)	45 (67.2)	37 (57.8)	44 (64.7)	48 (68.6)	45 (59.2)
All adverse drug reactions	9 (12.0)	10 (15.4)	16 (23.9)	9 (14.1)	16 (23.5)	13 (18.6)	13 (17.1)
Serious adverse events	3 (4.0)	0 (0.0)	1 (1.5)	1 (1.6)	1 (1.5)	1 (1.4)	1 (1.3)
Adverse events leading to treatment discontinuation	1 (1.3)	2 (3.1)	0 (0.0)	3 (4.7)	4 (5.9)	5 (7.1)	3 (3.9)
Hypoglycaemia	2 (2.7)	1 (1.5)	1 (1.5)	0 (0.0)	0 (0.0)	2 (2.9)	1 (1.3)
Pollakiuria	0 (0.0)	1 (1.5)	1 (1.5)	1 (1.6)	1 (1.5)	2 (2.9)	2 (2.6)
Nocturia	0 (0.0)	0 (0.0)	1 (1.5)	0 (0.0)	2 (2.9)	0 (0.0)	3 (3.9)
Urinary tract infection events	3 (4.0)	3 (4.6)	5 (7.5)	8 (12.5)	8 (11.8)	4 (5.7)	5 (6.6)
Genital events	1 (1.3)	5 (7.7)	6 (9.0)	5 (7.8)	3 (4.4)	9 (12.9)	2 (2.6)
Renal disorder	0 (0.0)	3 (4.6)	1 (1.5)	0 (0.0)	1 (1.5)	3 (4.3)	0 (0.0)

Adjusted mean [two-sided 95% CI]

Number of subjects with the event (incidence %)

PMDA considers as follows:

There is no problem with the proposed once daily oral administration of dapagliflozin. There is no problem with the unspecified timing of dosing for the following reasons: no substantial difference in the efficacy or safety was observed in the foreign clinical studies conducted using both morning dose and evening dose regimens although Japanese clinical studies exclusively used a morning dose regimen; no substantial difference was observed in urinary glucose excretion immediately after dosing under fasted conditions between dapagliflozin 2.5 mg and dapagliflozin 10 mg; and there was no food effect on AUC_{inf} . The above issues will be finalized, taking account of comments from the Expert Discussion.

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

The applicant explained as follows:

In a Japanese phase II study (Study D1692C00005), a statistically significant decrease in the primary endpoint of the change in HbA1c from baseline to Week 12 of treatment [Table 16] was

¹⁷⁰ Study MB102002: A placebo-controlled, double-blind, multiple dose escalation study in foreign healthy adult subjects that evaluated the safety, pharmacokinetics, and pharmacodynamic effects of once daily oral doses of dapagliflozin at 2.5, 10, 20, 50, or 100 mg for 14 days under fasted conditions.

observed in all dapagliflozin groups (1, 2.5, 5, 10 mg groups) compared with the placebo group. In addition, the decrease was greater in the 5 and 10 mg groups than in the 1 and 2.5 mg groups. Therefore, 5 and 10 mg were selected as dose levels for Japanese phase III studies. In the Japanese phase III study (Study D1692C00006), a statistically significant decrease in the primary endpoint of the change in HbA1c from baseline to Week 24 of treatment [Table 18] was observed in the 5 and 10 mg groups compared with the placebo group. It has been reported that the level of decrease in HbA1c in patients with diabetes mellitus after receiving hypoglycemic agents depends on baseline HbA1c (Bloomgarden ZT, et al. *Diabetes Care*. 2006;29:2137-9, DeFronzo RA, et al. *Diabet Med*. 2010;27:309-17), groups of subjects with higher baseline HbA1c tended to show a greater adjusted mean change in HbA1c in the Japanese and foreign clinical studies. In the Japanese phase III long-term treatment study (Study D1692C00012), in which the dose was allowed to be increased to 10 mg from a visit at or after Week 16 if HbA1c was >7.5% and there were no safety concerns at Week 12 of treatment or later, the proportion of subjects who received a dose increase was 18.5% in the monotherapy group, 44.3% in the dapagliflozin + SU group, 33.9% in the dapagliflozin + DPP-4 group, 19.7% in the dapagliflozin + α-GI group, 25.4% in the dapagliflozin + BG group, 26.6% in the dapagliflozin + TZD group, 26.5% in the dapagliflozin + glinide group, and 48.0% in the dapagliflozin + GLP-1 group. The proportion of subjects who showed a decrease in HbA1c by >0.3% at 16 weeks after the dose increase was 22.0% (11 of 50 subjects) in the monotherapy group, 30.2% (19 of 63 subjects) in the dapagliflozin + SU group, 57.1% (12 of 21 subjects) in the dapagliflozin + DPP-4 group, 20.0% (3 of 15 subjects) in the dapagliflozin + α-GI group, 15.0% (3 of 20 subjects) in the dapagliflozin + BG group, 15.8% (3 of 19 subjects) in the dapagliflozin + TZD group, 42.9% (6 of 14 subjects) in the dapagliflozin + glinide group, and 32.0% (8 of 25 subjects) in the dapagliflozin + GLP-1 group, demonstrating the effects of dose increase in spite of the variation of the degree of effects among concomitant drugs. Regarding safety, the incidences of adverse events among subjects with no dose increase and subjects with a dose increase were 78.9% (157 of 199 subjects) and 80.0% (40 of 50 subjects), respectively, in the monotherapy group; 72.9% (43 of 59 subjects) and 73.0% (46 of 63 subjects), respectively, in the dapagliflozin + SU group; 73.2% (30 of 41 subjects) and 81.0% (17 of 21 subjects), respectively, in the dapagliflozin + DPP-4 group; 60.0% (27 of 45 subjects) and 75.0% (12 of 16 subjects), respectively, in the dapagliflozin + α-GI group; 80.4% (41 of 51 subjects) and 75.0% (15 of 20 subjects), respectively, in the dapagliflozin + metformin group; 73.3% (33 of 45 subjects) and 63.2% (12 of 19 subjects), respectively, in the dapagliflozin + TZD group; 65.7% (23 of 35 subjects) and 78.6% (11 of 14 subjects), respectively, in the dapagliflozin + glinide group; and 72.0% (18 of 25 subjects) and 76.0% (19 of 25 subjects), respectively, in the dapagliflozin + GLP-1 group; showing no substantial difference between subjects with no dose increase and subjects with a dose increase. The incidence of adverse drug reactions among subjects with no dose increase and subjects with a dose increase were 24.6% (49 of 199 subjects) and 26.0% (13 of 50 subjects), respectively, in the monotherapy group; 22.0% (13 of 59 subjects) and 11.1% (7 of 63 subjects), respectively, in the dapagliflozin + SU group; 19.5% (8 of 41 subjects) and 14.3% (3 of 21 subjects), respectively, in the dapagliflozin + DPP-4 group; 11.1% (5 of 45 subjects) and 12.5% (2 of 16 subjects), respectively, in the dapagliflozin + α-GI group; 27.5% (14 of 51 subjects) and 30.0% (6 of 20 subjects), respectively, in the dapagliflozin + metformin group; 15.6% (7 of 45 subjects) and 5.3% (1 of 19 subjects), respectively, in the dapagliflozin + TZD group; 20.0% (7 of 35 subjects) and 21.4% (3 of 14 subjects), respectively, in the dapagliflozin + glinide group; and 16.0% (4 of 25 subjects) and 16.0% (4 of 25 subjects), respectively, in the dapagliflozin + GLP-1 group; showing no substantial difference between subjects with no dose increase and subjects with a dose increase. None of the subjects with a dose increase to 10 mg required a subsequent dose reduction leading to discontinuation. The incidence of dapagliflozin-related adverse events requiring attention was similar between subjects with no dose increase and subjects with a dose increase in all of the monotherapy and concomitant therapy groups, but the incidence of genital infection was higher among subjects with a dose increase than among subjects with no dose increase in all of the monotherapy and concomitant therapy groups. This difference was seen only among female subjects, and about half of genital infection-related

events reported by female subjects with a dose increase to 10 mg were already observed before the dose increase. In addition, the effects of dose increase of dapagliflozin in patients with renal impairment were evaluated by stratifying subjects with a dose increase into those with baseline eGFR ($\text{mL}/\text{min}/1.73 \text{ m}^2$) <60 and those with ≥ 60 and determining the proportion of subjects who showed a decrease in HbA1c at 16 weeks after the dose increase. The results showed that the proportions of subjects with baseline eGFR <60 and those with ≥ 60 who showed a decrease in HbA1c at 16 weeks after the dose increase were 66.7% (6 of 9 subjects) and 65.9% (27 of 41 subjects), respectively, in the monotherapy group, showing no substantial difference according to baseline eGFR. Also from a safety perspective, adverse events that were reported after the dose increase by subjects with baseline eGFR <60 were mostly mild in severity, and a causal relationship to the study drug was ruled out for the events.

Based on the above, the recommended dose of dapagliflozin in Japanese patients with type 2 diabetes mellitus would be 5 mg orally once daily, and further benefit could be expected by increasing the dose to 10 mg once daily with close monitoring of the patient if 5 mg is insufficient.

PMDA considers as follows:

There is no problem with the proposed usual dosage of dapagliflozin 5 mg once daily. There is no major problem in allowing a dose increase to 10 mg in subjects who are not adequately responsive to dapagliflozin 5 mg, because a certain proportion of subjects showed a decrease in HbA1c at 16 weeks after the dose increase in spite of the variation in the degree of effects of dose increase across concomitant therapies. The above issues will be finalized, taking account of comments from the Expert Discussion.

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

4.(iii).B.(6).1 Patients with renal impairment

The applicant explained as follows:

In a clinical pharmacology study (Foreign Study MB102007), the exposure (AUC) to dapagliflozin in patients with mild, moderate, and severe renal impairment was 28%, 52%, and 75% higher than that in patients with normal renal function, respectively. The daily urinary glucose excretion in patients with normal renal function was 84.9 g and those in patients with mild, moderate, and severe renal impairment were 51.8, 17.5, and 10.7 g, respectively, showing a greater decrease in glucose excretion in patients with severer renal impairment. The change in HbA1c by eGFR ($\text{mL}/\text{min}/1.73 \text{ m}^2$) in Japanese Study D1692C00006 was as shown in Table 28. The difference between the dapagliflozin 5 mg and placebo groups was similar between subjects with eGFR of ≥ 60 and <90 and subjects with eGFR of ≥ 45 and <60 , while the difference between the dapagliflozin 10 mg and placebo groups was smaller in subjects with eGFR of ≥ 45 and <60 than in subjects with eGFR of ≥ 60 and <90 . Each group included only 2 subjects with eGFR ≥ 90 .

Table 28. Change from baseline to Week 24 of treatment in HbA1c by eGFR in Study D1692C00006

eGFR ($\text{mL}/\text{min}/1.73 \text{ m}^2$)		Placebo group	Dapagliflozin 5 mg group	Dapagliflozin 10 mg group
≥ 45 and <60	Number of subjects	n = 24	n = 23	n = 24
	Baseline	7.34 ± 0.621	7.44 ± 0.532	7.55 ± 0.701
	Week 24 (LOCF)	7.30 ± 0.891	6.99 ± 0.480	7.20 ± 0.587
	Change from baseline	-0.10 [-0.32, 0.13]	-0.46 [-0.69, -0.23]	-0.31 [-0.53, -0.08]
	Between-group difference versus placebo	-	-0.37 [-0.68, -0.05]	-0.21 [-0.53, 0.10]
≥ 60 and <90	Number of subjects	n = 57	n = 61	n = 61
	Baseline	7.59 ± 0.625	7.52 ± 0.788	7.43 ± 0.579
	Week 24 (LOCF)	7.53 ± 0.827	7.13 ± 0.753	6.94 ± 0.497
	Change from baseline	-0.01 [-0.15, 0.14]	-0.37 [-0.51, -0.23]	-0.50 [-0.64, -0.36]
	Between-group difference versus placebo	-	-0.37 [-0.57, -0.16]	-0.49 [-0.70, -0.29]

Mean \pm SD, mean [two-sided 95% CI]

The change in HbA1c by eGFR in Japanese Study D1692C00012 was as shown in Table 29. Although some subgroups contained a limited number of subjects, HbA1c was generally decreased across all concomitant therapy groups.

Table 29. Change from baseline to Week 52 of treatment in HbA1c by eGFR in Study D1692C00012 (LOCF, excluding data from subjects after hyperglycaemia rescue therapy)

eGFR (mL/min/1.73 m ²)	Monotherapy group	Dapagliflozi n + SU group	Dapagliflozi n + DPP-4 group	Dapagliflozi n + α-GI group	Dapagliflozi n + BG group	Dapagliflozi n + TZD group	Dapagliflozi n + glinide group	Dapagliflozi n + GLP-1 group
≥45 and <60	-0.43 ± 0.852 (n = 61)	-0.50 ± 0.564 (n = 25)	-0.68 ± 0.475 (n = 13)	-0.72 ± 0.664 (n = 16)	0.10 ± 0.100 (n = 3)	-0.59 ± 0.727 (n = 12)	-0.55 ± 0.327 (n = 10)	-0.58 ± 0.647 (n = 16)
≥60 and <90	-0.73 ± 0.633 (n = 175)	-0.68 ± 0.673 (n = 88)	-0.57 ± 0.597 (n = 47)	-0.83 ± 0.717 (n = 39)	-0.61 ± 0.635 (n = 57)	-0.95 ± 0.768 (n = 49)	-0.79 ± 0.705 (n = 36)	-0.35 ± 0.797 (n = 31)
≥90	-0.86 ± 0.775 (n = 13)	-0.80 ± 1.227 (n = 9)	-0.60 ± 0.566 (n = 2)	-0.90 ± 0.374 (n = 6)	-1.06 ± 0.889 (n = 9)	-0.37 ± 0.153 (n = 3)	-1.03 ± 0.839 (n = 3)	-1.43 ± 1.150 (n = 3)

Mean ± SD

The efficacy in patients with moderate renal impairment was also evaluated in Foreign Study MB102029.¹⁷¹ No statistically significant differences in efficacy results were observed between the placebo and dapagliflozin groups, as shown in Table 30. An evaluation by eGFR (≥30 and <45 versus ≥45 and <60) suggested lack of efficacy in subjects with eGFR of ≥30 and <45, although a clinically meaningful decrease was observed in subjects with eGFR of ≥45 and <60 in the dapagliflozin groups.

Table 30. Change from baseline to Week 24 of treatment in HbA1c by eGFR in Study MB102029 (excluding data from subjects after hyperglycaemia rescue therapy)

eGFR (mL/min/1.73 m ²)		Placebo group	Dapagliflozin 5 mg group	Dapagliflozin 10 mg group
≥30 and <60	Number of subjects	n = 82	n = 83	n = 82
	Baseline	8.53 ± 1.285	8.30 ± 1.040	8.22 ± 0.973
	Week 24 (LOCF)	8.18 ± 1.204	7.97 ± 1.150	7.90 ± 0.930
	Change from baseline	-0.32 [-0.66, 0.01]	-0.41 [-0.74, -0.07]	-0.44 [-0.77, -0.10]
	Between-group difference versus placebo	-	-0.08 [-0.37, 0.20]	-0.11 [-0.40, 0.17]
≥30 and <45	Number of subjects	n = 33	n = 41	n = 45
	Baseline	8.23 ± 1.197	8.49 ± 1.157	8.12 ± 1.001
	Week 24 (LOCF)	7.79 ± 1.149	7.97 ± 1.250	7.78 ± 0.864
	Change from baseline	-0.52 [-1.08, 0.03]	-0.47 [-1.01, 0.06]	-0.45 [-0.96, 0.05]
	Between-group difference versus placebo	-	0.05 [-0.37, 0.47]	0.07 [-0.34, 0.48]
≥45 and <60	Number of subjects	n = 40	n = 35	n = 32
	Baseline	8.78 ± 1.318	8.13 ± 0.928	8.25 ± 0.892
	Week 24 (LOCF)	8.62 ± 1.201	7.93 ± 1.086	8.03 ± 1.002
	Change from baseline	-0.11 [-0.57, 0.35]	-0.47 [-0.97, 0.02]	-0.44 [-0.94, 0.07]
	Between-group difference versus placebo	-	-0.37 [-0.83, 0.10]	-0.33 [-0.80, 0.14]

Mean ± SD, adjusted mean [two-sided 95% CI]

Regarding safety, the incidences of adverse events by eGFR in the monotherapy group in Japanese Studies D1692C00006 and D1692C00012 were as shown in Table 31. In Japanese Study

¹⁷¹ Foreign Study MB102029: A placebo-controlled, randomized, double-blind, parallel-group phase II/III study in type 2 diabetes mellitus patients with moderate renal impairment that evaluated the efficacy and safety of dapagliflozin 5 and 10 mg (consisting of a 24-week confirmatory study and subsequent long-term extension periods [28-week + 52-week], for a total duration of 104 weeks).

D1692C00006, only 5 subjects in the placebo group, 2 subjects in the dapagliflozin 5 mg group, and 2 subjects in the dapagliflozin 10 mg group had eGFR of ≥ 90 . In the main Japanese and foreign clinical studies, treatment discontinuation criteria for high serum creatinine values were established in order to discontinue study treatment in patients who reported abnormal renal function tests or renal failure specified by the criteria. Japanese clinical studies (excluding the dapagliflozin + BG group) included patients with eGFR (mL/min/1.73 m²) ≥ 45 at study entry, and treatment was to be discontinued in patients who reported eGFR < 45 after the start of treatment by considering the decrease as an adverse event. In Japanese Study D1692C00006, the incidence of renal disorder/renal failure-related events was higher in patients with moderate renal impairment (eGFR of ≥ 30 and < 60) than in patients with mild renal impairment (eGFR of ≥ 60 and < 90) in both the placebo and dapagliflozin groups. Similarly, among dapagliflozin-related events requiring attention observed in the monotherapy group in Japanese Study D1692C00012, the incidence of renal disorder/renal failure-related events was higher in patients with moderate renal impairment (eGFR of ≥ 30 and < 60) than in patients with mild renal impairment (eGFR of ≥ 60 and < 90). An analysis according to the concomitant drug showed no trend toward differences in incidence across monotherapy and concomitant therapy groups.

Table 31. Incidence of adverse events by eGFR during monotherapy in 2 Japanese studies

	Japanese Study D1692C00006						Japanese Study D1692C00012		
	eGFR ≥ 30 and < 60			eGFR ≥ 60 and < 90			eGFR ≥ 30 and < 60	eGFR ≥ 60 and < 90	eGFR ≥ 90
	Placebo group (n = 24)	5 mg group (n = 23)	10 mg group (n = 25)	Placebo group (n = 58)	5 mg group (n = 61)	10 mg group (n = 61)	Monotherapy group (n = 61)	Monotherapy group (n = 175)	Monotherapy group (n = 13)
All adverse events	16 (66.7)	8 (34.8)	19 (76.0)	29 (50.0)	33 (54.1)	37 (60.7)	51 (83.6)	133 (76.0)	13 (100.0)
All adverse drug reactions	5 (20.8)	2 (8.7)	4 (16.0)	7 (12.1)	4 (6.6)	12 (19.7)	17 (27.9)	41 (23.4)	4 (30.8)
Serious adverse events	1 (4.2)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.6)	2 (3.3)	12 (6.9)	0 (0.0)
Adverse events leading to treatment discontinuation	4 (16.7)	2 (8.7)	6 (24.0)	1 (1.7)	1 (1.6)	1 (1.6)	6 (9.8)	9 (5.1)	0 (0.0)
Hypoglycaemia	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	2 (3.3)	0 (0.0)	6 (3.4)	0 (0.0)
Pollakiuria	0 (0.0)	0 (0.0)	0 (0.0)	2 (3.4)	2 (3.3)	5 (8.2)	1 (1.6)	11 (6.3)	2 (15.4)
Urinary tract infection events	1 (4.2)	0 (0.0)	1 (4.0)	1 (1.7)	0 (0.0)	1 (1.6)	3 (4.9)	6 (3.4)	0 (0.0)
Genital events	0 (0.0)	1 (4.3)	1 (4.0)	1 (1.7)	0 (0.0)	1 (1.6)	3 (4.9)	7 (4.0)	0 (0.0)
Renal disorder/renal failure	3 (12.5)	2 (8.7)	6 (24.0)	0 (0.0)	0 (0.0)	0 (0.0)	5 (8.2)	0 (0.0)	0 (0.0)
Volume depletion	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.6)	1 (0.6)	1 (7.7)
Fracture	0 (0.0)	0 (0.0)	0 (0.0)	1 (1.7)	1 (1.6)	2 (3.3)	0 (0.0)	4 (2.3)	2 (15.4)

Number of subjects with event (incidence %)

The incidence of adverse events by eGFR in 30-MU (short-term) and 30-MU (short-term + long-term) based on updated data from a pooled analysis (30-MU¹⁵³) was as shown in Table 32. In both 30-MU (short-term) and 30-MU (short-term + long-term), among dapagliflozin-related events requiring attention, the incidence of renal disorder/renal failure-related events was high in patients with eGFR of ≥ 45 and < 60 in both the placebo and dapagliflozin 10 mg groups, and higher in the dapagliflozin group than in the placebo group. Additionally, adverse events related to volume depletion also increased. In 30-MU (short-term), eGFR did not change throughout the 24-week treatment period in the placebo group. However, in the dapagliflozin 10 mg group, the change (mean \pm SE) from baseline in eGFR decreased at Week 1 (-1.482 ± 0.75987 mL/min/1.73 m² [n = 92]) and then increased to near baseline at Week 4 (-0.152 ± 0.51327 mL/min/1.73 m² [n = 242]) in subjects with eGFR of ≥ 30 and < 60 . Among subjects with eGFR ≥ 60 , the change decreased at Week 1 (-4.42 ± 0.32342 mL/min/1.73 m² [n = 1010]) and then gradually increased from Week 4 (-3.064 ± 0.26463 mL/min/1.73 m² [n = 1819]) at Week 4, -2.287 ± 0.27012

mL/min/1.73 m² [n = 1776] at Week 8, -1.130 ± 0.32967 mL/min/1.73 m² [n = 1159] at Week 12, -1.887 ± 0.27959 mL/min/1.73 m² [n = 1754] at Week 24).

Table 32. Incidence of adverse events by eGFR in 30-MU

		eGFR ≥45 and <60		eGFR ≥60 and <90		eGFR ≥90	
30-MU (Short-term)		Placebo group (n = 238)	Dapagliflozin 10 mg group (n = 222)	Placebo group (n = 1281)	Dapagliflozin 10 mg group (n = 1303)	Placebo group (n = 744)	Dapagliflozin 10 mg group (n = 791)
		All adverse events	160 (67.2)	150 (67.6)	699 (54.6)	765 (58.7)	397 (53.4)
		All adverse drug reactions	41 (17.2)	60 (27.0)	142 (11.1)	224 (17.2)	73 (9.8)
		Serious adverse events	19 (8.0)	15 (6.8)	73 (5.7)	67 (5.1)	27 (3.6)
		Adverse events leading to treatment discontinuation	21 (8.8)	24 (10.8)	41 (3.2)	45 (3.5)	15 (2.0)
		Hypoglycaemia	51 (21.4)	45 (20.3)	178 (13.9)	186 (14.3)	48 (6.5)
		Pollakiuria	2 (0.8)	11 (5.0)	19 (1.5)	47 (3.6)	5 (0.7)
		Urinary tract infection events	13 (5.5)	15 (6.8)	42 (3.3)	60 (4.6)	23 (3.1)
		Genital events	1 (0.4)	15 (6.8)	4 (0.3)	72 (5.5)	9 (1.2)
		Renal disorder/renal failure	21 (8.8)	35 (15.8)	14 (1.1)	22 (1.7)	3 (0.4)
		Body fluid volume decreased	4 (1.7)	4 (1.8)	9 (0.7)	20 (1.5)	4 (0.5)
		Fracture	1 (0.4)	0 (0.0)	13 (1.0)	4 (0.3)	2 (0.3)
30-MU (Short-term + long-term)		Placebo group (n = 220)	Dapagliflozin 10 mg group (n = 208)	Placebo group (n = 1113)	Dapagliflozin 10 mg group (n = 1140)	Placebo group (n = 592)	Dapagliflozin 10 mg group (n = 634)
		All adverse events	180 (81.8)	168 (80.8)	779 (70.0)	843 (73.9)	416 (70.3)
		All adverse drug reactions	51 (23.2)	65 (31.3)	158 (14.2)	242 (21.2)	75 (12.7)
		Serious adverse events	44 (20.0)	34 (16.3)	167 (15.0)	165 (14.5)	66 (11.1)
		Adverse events leading to treatment discontinuation	36 (16.4)	47 (22.6)	78 (7.0)	75 (6.6)	22 (3.7)
		Hypoglycaemia	70 (31.8)	62 (29.8)	270 (24.3)	247 (21.7)	86 (14.5)
		Pollakiuria	4 (1.8)	10 (4.8)	17 (1.5)	47 (4.1)	6 (1.0)
		Urinary tract infection events	19 (8.6)	17 (8.2)	66 (5.9)	103 (9.0)	32 (5.4)
		Genital events	1 (0.5)	18 (8.7)	9 (0.8)	87 (7.6)	9 (1.5)
		Renal disorder/renal failure	33 (15.0)	53 (25.5)	36 (3.2)	53 (4.6)	6 (1.0)
		Body fluid volume decreased	6 (2.7)	7 (3.4)	16 (1.4)	26 (2.3)	5 (0.8)
		Fracture	3 (1.4)	1 (0.5)	21 (1.9)	15 (1.3)	6 (1.0)
		Number of subjects with the event (incidence %)					

The safety in patients with moderate renal impairment was evaluated also in Foreign Study MB102029.¹⁷¹ The safety results by eGFR (≥ 30 to <45 versus ≥ 45 to <60) were as shown in Table 33. Among dapagliflozin-related events requiring attention, the incidence of renal disorder/renal failure-related adverse events was high in patients with eGFR of ≥ 30 and <45 in both the placebo and dapagliflozin 10 mg groups, and higher in the dapagliflozin group than in the placebo group. Fracture was not reported in the placebo group, but reported by 7 subjects in the dapagliflozin 10 mg group, including 5 subjects (10.6%) with eGFR of ≥ 30 and <45 and 2 subjects (6.1%) with eGFR of ≥ 45 and <60 , showing a higher incidence in subjects with eGFR of ≥ 30 and <45 . Comparison of subject characteristics between Study MB102029 and 30-MU showed that serum P and parathyroid hormone levels at baseline were higher in Study MB102029 than in 30-MU, and the mean increases from baseline in serum P and parathyroid hormone levels in the

dapagliflozin group were higher in Study MB102029 than in 30-MU. The higher incidence of fracture in subjects with eGFR of ≥ 30 and < 45 is considered related to the increased serum P and parathyroid hormone levels in this population, which is at risk of secondary hyperparathyroidism and renal osteodystrophy. However, an analysis of incidence of fracture in subjects with eGFR ≥ 45 by renal function using data from 30-MU (short-term + long-term) revealed no difference from placebo in the incidence of fracture [Table 32]. In addition, a subgroup analysis of 30-MU (short-term) by eGFR (≥ 45 and < 60 versus ≥ 60 and < 90 versus ≥ 90) revealed no difference in the change from baseline depending on eGFR for any marker of bone formation and resorption. Therefore, dapagliflozin would not have a particularly large impact on bone metabolism in the population with baseline eGFR ≥ 45 .

**Table 33. Incidence of adverse events by eGFR in Foreign Study MB102029
(during 52 weeks of treatment)**

Event term	eGFR ≥ 30 and < 45		eGFR ≥ 45 and < 60	
	Placebo group (n = 34)	Dapagliflozin 10 mg group (n = 47)	Placebo group (n = 41)	Dapagliflozin 10 mg group (n = 33)
All adverse events	28 (82.4)	39 (83.0)	36 (87.8)	30 (90.9)
All adverse drug reactions	15 (44.1)	20 (42.6)	16 (39.0)	14 (42.4)
Serious adverse events	6 (17.6)	12 (25.5)	10 (24.4)	5 (15.2)
Adverse events leading to treatment discontinuation	8 (23.5)	4 (8.5)	8 (19.5)	3 (9.1)
Hypoglycaemia	13 (38.2)	16 (34.0)	22 (53.7)	13 (39.4)
Pollakiuria	0 (0.0)	4 (8.5)	3 (7.3)	6 (18.2)
Urinary tract infection events	3 (8.8)	5 (10.6)	4 (9.8)	2 (6.1)
Genital events	1 (2.9)	2 (4.3)	2 (4.9)	4 (12.1)
Renal disorder/renal failure	1 (2.9)	3 (6.4)	1 (2.4)	1 (3.0)
Body fluid volume decreased	3 (8.8)	4 (8.5)	0 (0.0)	5 (15.2)
Fracture	0 (0.0)	5 (10.6)	0 (0.0)	2 (6.1)

Number of subjects with the event (incidence %)

Based on the above, while there would be no problem with efficacy and safety in patients with eGFR ≥ 45 , no efficacy would be available and the safety risk would be increased in patients with eGFR ≥ 30 and < 45 . Therefore, use of dapagliflozin in patients with eGFR ≥ 30 and < 45 is not recommended. Use of dapagliflozin in patients with eGFR < 30 is not recommended because these patients were excluded from the clinical trials and no benefit can be expected.

PMDA considers as follows:

From an efficacy perspective, urinary glucose excretion after administration of dapagliflozin was decreased depending on the severity of renal impairment, and Foreign Study MB102029 conducted in patients with moderate renal impairment with eGFR ≥ 30 and < 60 failed to confirm the efficacy of dapagliflozin, although a decrease in HbA1c was observed in patients with baseline eGFR ≥ 45 in Japanese clinical studies. Therefore, it is necessary to provide information to the effect that the efficacy of dapagliflozin is diminished with decreasing renal function, because adequate efficacy cannot be expected from dapagliflozin in type 2 diabetic patients with renal impairment of moderate or greater severity. From a safety perspective, PMDA accepted the applicant's explanation that there was no clear trend toward an increased risk in patients with eGFR ≥ 45 . However, effects of continued treatment with dapagliflozin in patients who showed a decreased eGFR < 45 have not been adequately evaluated because treatment was planned to be discontinued in subjects in whom eGFR was decreased to < 45 , and there was a trend toward an increased incidence of fracture in patients with eGFR ≥ 30 and < 45 during a limited treatment duration in Foreign Study MB102029. In addition, a pooled analysis showed a trend toward increased incidences of adverse events related to volume depletion and renal disorder in the subgroup of subjects with eGFR ≥ 45 and < 60 . Given the foregoing, the appropriateness of the use of dapagliflozin in patients with renal impairment of moderate or greater severity should be carefully considered in terms of efficacy and safety. Given the limited number of subjects included in the clinical studies, it is necessary to continue to collect information on the safety and

efficacy in patients with renal impairment via post-marketing surveillance. The above issues will be finalized, taking account of comments from the Expert Discussion.

4.(iii).B.(6).2 Patients with hepatic impairment

The applicant explained as follows:

In a clinical pharmacology study (Foreign Study MB102027), C_{max} and AUC of dapagliflozin were 12% and 36%, respectively, higher in patients with moderate hepatic impairment, and 40% and 67%, respectively, higher in patients with severe hepatic impairment than in healthy subjects. Patients with severe hepatic dysfunction and/or markedly abnormal liver-function tests (AST and/or ALT >3-fold the upper limit of normal, total bilirubin >2.0 mg/dL) were excluded from the Japanese and foreign clinical studies. The incidences of adverse events in the monotherapy group in Japanese Studies D1692C00006 and D1692C00012 by hepatic impairment (AST/ALT \leq normal ranges, AST/ALT >the upper limit of normal) were as shown in Table 34. Data from Japanese Study D1692C00006 showed no substantial difference in the incidence of adverse events in a subgroup of subjects with hepatic impairment between the dapagliflozin and placebo groups, although the number of subjects studied was limited. In addition, adverse events leading to study treatment discontinuation or serious adverse events were reported by a small number of subjects. Similarly, in the monotherapy group in Japanese Study D1692C00012, the incidence of dapagliflozin-related adverse events did not increase in the subgroup of subjects with hepatic impairment, although the number of such subjects was limited. In addition, adverse events leading to study drug discontinuation or serious adverse events were reported by a small number of subjects. An analysis according to the concomitant drug showed no trend toward differences in incidence across monotherapy and concomitant therapy groups.

Table 34. Incidence of adverse events by hepatic impairment during monotherapy in 2 Japanese studies

	Japanese Study D1692C00006						Japanese Study D1692C00012	
	Subjects without hepatic impairment			Subjects with hepatic impairment			Subjects without hepatic impairment	Subjects with hepatic impairment
	Placebo group (n = 70)	5 mg group (n = 74)	10 mg group (n = 79)	Placebo group (n = 17)	5 mg group (n = 12)	10 mg group (n = 9)	Monotherapy group (n = 210)	Monotherapy group (n = 39)
All adverse events	38 (54.3)	34 (45.9)	53 (67.1)	7 (41.2)	7 (58.3)	4 (44.4)	167 (79.5)	30 (76.9)
Serious adverse events	0 (0.0)	0 (0.0)	1 (1.3)	1 (5.9)	0 (0.0)	0 (0.0)	14 (6.7)	0 (0.0)
Adverse events leading to treatment discontinuation	2 (2.9)	2 (2.7)	7 (8.9)	3 (17.6)	1 (8.3)	0 (0.0)	14 (6.7)	1 (2.6)
SOC “Hepatobiliary disorders”	0 (0.0)	0 (0.0)	1 (1.3)	0 (0.0)	1 (8.3)	0 (0.0)	2 (1.0)	2 (5.1)

Number of subjects with the event (incidence %)

Using updated data from a pooled analysis (30-MU¹⁵³), a comparative analysis was made between subjects with normal baseline ALT and/or AST (\leq 1-fold the upper limit of normal) and subjects with high baseline ALT and/or AST (>1-fold the upper limit of normal). The incidence of adverse events in subjects with normal liver-function tests was 54.9% (1194 of 2174 subjects) and 59.3% (1187 of 2003 subjects) in the placebo and dapagliflozin 10 mg groups, respectively. The incidence of adverse events in subjects with high liver-function tests was 62.8% (245 of 390 subjects) and 64.1% (229 of 357 subjects), respectively. The incidence of adverse drug reactions in subjects with normal liver-function tests was 11.2% (244 of 2174 subjects) and 16.7% (335 of 2003 subjects) in the placebo and dapagliflozin 10 mg groups, respectively. The incidence of adverse drug reactions in subjects with high liver-function tests was 14.1% (55 of 390 subjects) and 20.7% (74 of 357 subjects), respectively. In the dapagliflozin 10 mg group, the incidences were higher in subjects with high liver-function tests than in subjects with normal liver-function tests, but a similar trend was observed also in the placebo group.

PMDA accepted the applicant's explanation. However, it is necessary to continue to collect information on the safety in patients with hepatic impairment via post-marketing surveillance because dapagliflozin has not been studied in clinical studies in patients with severe hepatic impairment.

4.(iii).B.(6).3 Elderly patients

The applicant explained as follows:

In population pharmacokinetic analyses and a pooled analysis from clinical pharmacology studies in foreign subjects, effects of age on the exposure were evaluated. The results showed no correlation between AUC and age. The incidences of adverse events by age in the monotherapy group in Japanese Studies D1692C00006 and D1692C00012 were as shown in Table 35. In Japanese Study D1692C00006, only 3 subjects in the placebo group, 4 subjects in the dapagliflozin 5 mg group, and 1 subject in the dapagliflozin 10 mg group were aged ≥ 75 years. In the monotherapy group in Japanese Study D1692C00012, among dapagliflozin-related events requiring attention, the incidences of events related to volume depletion, urinary tract infection, and renal disorder/renal failure were higher in subjects aged ≥ 65 years than in subjects aged < 65 years. An analysis by concomitant drug showed no trend toward differences in incidence across monotherapy and concomitant therapy groups, although incidence of some of these events was low.

Table 35. Incidence of adverse events by age during monotherapy in 2 Japanese studies

	Japanese Study D1692C00006						Japanese Study D1692C00012		
	<65 years of age			≥ 65 years of age ^{a)}			<65 years of age	≥ 65 years of age ^{a)}	≥ 75 years of age
	Placebo group (n = 55)	5 mg group (n = 59)	10 mg group (n = 67)	Placebo group (n = 32)	5 mg group (n = 27)	10 mg group (n = 21)	Monotherapy group (n = 182)	Monotherapy group (n = 67)	Monotherapy group (n = 12)
All adverse events	28 (50.9)	25 (42.4)	43 (64.2)	17 (53.1)	16 (59.3)	14 (66.7)	143 (78.6)	54 (80.6)	8 (66.7)
All adverse drug reactions	6 (10.9)	3 (5.1)	14 (20.9)	6 (18.8)	3 (11.1)	3 (14.3)	41 (22.5)	21 (31.3)	2 (16.7)
Serious adverse events	0 (0.0)	0 (0.0)	0 (0.0)	1 (3.1)	0 (0.0)	1 (4.8)	7 (3.8)	7 (10.4)	2 (16.7)
Adverse events leading to treatment discontinuation	2 (3.6)	2 (3.4)	3 (4.5)	3 (9.4)	1 (3.7)	4 (19.0)	9 (4.9)	6 (9.0)	1 (8.3)
Hypoglycaemia	0 (0.0)	0 (0.0)	2 (3.0)	0 (0.0)	0 (0.0)	0 (0.0)	5 (2.7)	1 (1.5)	0 (0.0)
Pollakiuria	1 (1.8)	1 (1.7)	4 (6.0)	1 (3.1)	1 (3.7)	1 (4.8)	12 (6.6)	2 (3.0)	0 (0.0)
Urinary tract infection events	1 (1.8)	0 (0.0)	1 (1.5)	1 (3.1)	0 (0.0)	1 (4.8)	4 (2.2)	5 (7.5)	0 (0.0)
Genital events	0 (0.0)	0 (0.0)	2 (3.0)	1 (3.1)	1 (3.7)	0 (0.0)	8 (4.4)	2 (3.0)	0 (0.0)
Renal disorder/renal failure	0 (0.0)	1 (1.7)	3 (4.5)	3 (9.4)	1 (3.7)	3 (14.3)	3 (1.6)	2 (3.0)	1 (8.3)
Body fluid volume decreased	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	0 (0.0)	1 (0.5)	2 (3.0)	0 (0.0)
Fracture	1 (1.8)	0 (0.0)	1 (1.5)	0 (0.0)	1 (3.7)	1 (4.8)	6 (3.3)	0 (0.0)	0 (0.0)

Number of subjects with the event (incidence %)

a) Including subjects aged ≥ 75 years

The incidence of adverse events by age based on updated data from a pooled analysis (30-MU¹⁵³) was as shown in Table 36. In both 30-MU (short-term) and 30-MU (short-term + long-term), among dapagliflozin-related events requiring attention, the incidence of renal disorder/renal failure-related adverse events was higher in subjects aged ≥ 65 years in both the placebo and dapagliflozin 10 mg groups, and higher in the dapagliflozin 10 mg group than in the placebo group. In 30-MU (short-term), renal disorder/renal failure-related adverse events reported by $> 1\%$ of subjects aged ≥ 65 years in the dapagliflozin 10 mg group include creatinine renal clearance

decreased (1.8% in the placebo group, 3.0% in the dapagliflozin 10 mg group), renal impairment (0.8% in the placebo group, 2.3% in the dapagliflozin 10 mg group), and blood creatinine increased (0.4% in the placebo group, 1.1% in the dapagliflozin 10 mg group). In subjects aged ≥65 years, only a few reported renal failure (4 subjects in the dapagliflozin 10 mg group, 3 subjects in the placebo group). In the dapagliflozin group in 30-MU (short-term) and 30-MU (short-term + long-term), the incidence of adverse events related to volume depletion was higher in subjects aged ≥65 years, who appear to be relatively susceptible to diuretic effect, than in subjects aged <65 years.

Table 36. Incidence of adverse events by age in 30-MU

		<65 years of age		≥65 years of age ^{a)}		≥75 years of age	
30-MU (Short-term)		Placebo group (n = 1584)	Dapagliflozin 10 mg group (n = 1695)	Placebo group (n = 711)	Dapagliflozin 10 mg group (n = 665)	Placebo group (n = 81)	Dapagliflozin 10 mg group (n = 98)
	All adverse events	877 (55.4)	1017 (60.0)	402 (56.5)	399 (60.0)	49 (60.5)	63 (64.3)
	All adverse drug reactions	180 (11.4)	271 (16.0)	81 (11.4)	138 (20.8)	11 (13.6)	24 (24.5)
	Serious adverse events	66 (4.2)	66 (3.9)	57 (8.0)	54 (8.1)	6 (7.4)	7 (7.1)
	Adverse events leading to treatment discontinuation	41 (2.6)	50 (2.9)	41 (5.8)	52 (7.8)	7 (8.6)	13 (13.3)
	Hypoglycaemia	181 (11.4)	215 (12.7)	103 (14.5)	109 (16.4)	9 (11.1)	13 (13.3)
	Pollakiuria	21 (1.3)	53 (3.1)	6 (0.8)	25 (3.8)	0 (0.0)	2 (2.0)
	Urinary tract infection events	51 (3.2)	77 (4.5)	30 (4.2)	33 (5.0)	5 (6.2)	4 (4.1)
	Genital events	11 (0.7)	97 (5.7)	3 (0.4)	33 (5.0)	0 (0.0)	6 (6.1)
	Renal disorder/renal failure	15 (0.9)	25 (1.5)	27 (3.8)	51 (7.7)	6 (7.4)	13 (13.3)
30-MU (Short-term + long-term)	Body fluid volume decreased	11 (0.7)	16 (0.9)	6 (0.8)	11 (1.7)	1 (1.2)	3 (3.1)
	Fracture	8 (0.5)	6 (0.4)	9 (1.3)	2 (0.3)	2 (2.5)	0 (0.0)
		Placebo group (n = 1301)	Dapagliflozin 10 mg group (n = 1406)	Placebo group (n = 655)	Dapagliflozin 10 mg group (n = 620)	Placebo group (n = 77)	Dapagliflozin 10 mg group (n = 97)
	All adverse events	920 (70.7)	1028 (73.1)	479 (73.1)	480 (77.4)	58 (75.3)	78 (80.4)
	All adverse drug reactions	189 (14.5)	286 (20.3)	102 (15.6)	162 (26.1)	13 (16.9)	30 (30.9)
	Serious adverse events	154 (11.8)	154 (11.0)	132 (20.2)	124 (20.0)	14 (18.2)	19 (19.6)
	Adverse events leading to treatment discontinuation	65 (5.0)	83 (5.9)	80 (12.2)	89 (14.4)	17 (22.1)	26 (26.8)
	Hypoglycaemia	273 (21.0)	290 (20.6)	164 (25.0)	145 (23.4)	17 (22.1)	19 (19.6)
	Pollakiuria	21 (1.6)	53 (3.8)	7 (1.1)	26 (4.2)	0 (0.0)	3 (3.1)
	Urinary tract infection events	71 (5.5)	124 (8.8)	50 (7.6)	50 (8.1)	7 (9.1)	8 (8.2)
	Genital events	13 (1.0)	115 (8.2)	6 (0.9)	41 (6.6)	0 (0.0)	7 (7.2)
	Renal disorder/renal failure	30 (2.3)	49 (3.5)	52 (7.9)	87 (14.0)	16 (20.8)	29 (29.9)
	Body fluid volume decreased	16 (1.2)	24 (1.7)	11 (1.7)	14 (2.3)	2 (2.6)	3 (3.1)
	Fracture	14 (1.1)	16 (1.1)	18 (2.7)	7 (1.1)	2 (2.6)	1 (1.0)

Number of subjects with the event (incidence %)

a) Including subjects aged ≥75 years

PMDA considers as follows:

Although there was a trend toward a higher incidence of events related to volume depletion, urinary tract infection, and renal disorder/renal failure in subjects aged ≥65 years in Japanese clinical studies as well as a trend toward a higher incidence of events related to volume depletion and renal disorder/renal failure in the dapagliflozin group compared with the placebo group

among subjects aged ≥65 years in a pooled analysis from foreign studies, there is no major problem on the premise that appropriate caution statements are provided. It is necessary to continue to collect information on the safety in elderly patients via post-marketing surveillance.

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

The applicant plans to conduct a post-marketing surveillance with a sample size of [REDACTED] and an observation period of [REDACTED] years to evaluate the long-term safety and efficacy of dapagliflozin. This surveillance will investigate genital infection- and urinary tract infection-related events as a priority.

PMDA considers as follows:

It is also necessary to collect information on the impact of concomitant drugs on the safety depending on their types and doses; adverse events related to urinary tract and genital infections; adverse events related to pollakiuria and polyuria; adverse events related to increased ketone bodies; safety regarding bone metabolism, renal impairment, and tumorigenesis; and safety and efficacy in patients with renal or hepatic impairment and elderly patients, etc. Appropriate safety measures should be taken from the following standpoints: dapagliflozin is a drug with a novel mechanism of action and has been recently approved overseas; the incidence of adverse events associated with volume depletion may be increased by the external environment such as seasons as compared with that under clinical trial settings; and urinary tract infections, which may progress in severity unless detected early, may be detected later in routine clinical settings than in clinical trial settings. Details of the post-marketing surveillance 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

Described in Review Report (2).

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

Described in Review Report (2).

IV. Overall Evaluation

Based on the submitted data, it is concluded that the efficacy of dapagliflozin in patients with type 2 diabetes mellitus has been demonstrated and its safety is considered acceptable in view of its observed benefits. The product is an oral hypoglycemic agent with a novel mechanism of action that offers a new therapeutic option for type 2 diabetes mellitus. PMDA considers that further investigation is necessary for the following issues: appropriateness of the use of dapagliflozin in patients with renal impairment of moderate or greater severity; impact of concomitant oral hypoglycemic agents on the safety depending on their types and doses; safety in hypoglycaemia, urinary tract infections, genital infections, polyuria/pollakiuria, volume depletion, increase in ketone body, weight decreased, renal disorder, bone metabolism, cardiovascular risks, and malignant tumors; safety in patients with renal or hepatic impairment; and safety in elderly patients.

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

Review Report (2)

December 24, 2013

I. Product Submitted for Registration

[Brand name]	Forxiga 5 mg tablets, Forxiga 10 mg tablets
[Non-proprietary name]	Dapagliflozin Propylene Glycolate Hydrate
[Applicant]	Bristol-Myers K.K.
[Date of application]	March 15, 2013

II. Content of the Review

The outline of the comments from the Expert Discussion and the subsequent review by the Pharmaceuticals and Medical Devices Agency (PMDA) is described in the following sections. 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) Efficacy of monotherapy

PMDA concluded that the efficacy of monotherapy has been demonstrated based on the results of a Japanese phase III study (Study D1692C00006) and the Japanese phase III long-term treatment study (Study D1692C00012), etc.

The above conclusion of PMDA was supported by the expert advisors.

2) Efficacy of concomitant therapies

PMDA concluded that the efficacy of each concomitant therapy has been confirmed based on the results of the Japanese phase III long-term treatment study (Study D1692C00012).

The above conclusion of PMDA was supported by the expert advisors.

(2) Safety

PMDA considers as follows:

Given the incidences of adverse events and adverse drug reactions with monotherapy and each concomitant therapy, the safety of dapagliflozin is acceptable on the premise that appropriate caution statements are provided. In addition, based on events of special interest for safety assessment (e.g., hypoglycaemia, polyuria/pollakiuria-related events, urinary tract infections, genital infections, volume depletion), it is necessary to continue to collect information via post-marketing surveillance, although no major problems have been found at present. Furthermore, it is also necessary to continue to collect information via post-marketing surveillance on the impact of concomitant oral hypoglycemic agents on the safety depending on their doses and types.

The above conclusion of PMDA was supported by the expert advisors.

Based on the above, PMDA instructed the applicant to take actions, and confirmed that appropriate actions have been taken regarding the description of caution statements in the package insert [see “(6) Draft risk management plan” for post-marketing investigations].

(3) Indication

PMDA concluded that there is no problem with the indication of “type 2 diabetes mellitus,” because the efficacy of monotherapy and concomitant therapies has been demonstrated through clinical studies conforming to OAD Guideline and their safety is acceptable. The above conclusion of PMDA was supported by the expert advisors.

(4) Dosage and administration

PMDA considers as follows:

There is no problem with once daily oral administration. There is no problem with the unspecified timing of dosing for the following reasons: no substantial difference in the efficacy or safety was observed in the foreign clinical studies conducted using both morning and evening dose regimens; no substantial difference was observed in urinary glucose excretion immediately after dosing under fasted conditions between 2.5 mg and 10 mg of dapagliflozin; and there was no food effect on AUC_{inf}. Regarding dose level, there is no problem with the usual dosage of dapagliflozin 5 mg once daily, and in addition, there is no major problem in allowing a dose increase to 10 mg in subjects who are not adequately responsive to dapagliflozin 5 mg.

The above conclusion of PMDA was supported by the expert advisors.

(5) Special populations

1) Patients with renal impairment

PMDA considers as follows:

From an efficacy perspective, it is necessary to provide information on the diminishing efficacy of dapagliflozin associated with severity of renal impairment, because adequate efficacy cannot be expected from dapagliflozin in type 2 diabetic patients with renal impairment of moderate or greater severity. From a safety perspective, the applicant’s explanation that there was no clear trend toward an increased risk in patients with baseline eGFR ≥ 45 is acceptable, but it is necessary to advise that the appropriateness of the use of dapagliflozin in patients with renal impairment of moderate or greater severity be carefully considered, because a pooled analysis showed a trend toward increased incidences of adverse events related to volume depletion and renal disorder in the subgroup of subjects with eGFR of ≥ 45 and < 60 . In addition, given the limited number of subjects in the clinical studies, it is necessary to continue to collect information on the impact of dapagliflozin on renal function and the safety and efficacy in patients with renal impairment via post-marketing surveillance.

The above conclusion of PMDA was supported by the expert advisors.

Based on the above, PMDA instructed the applicant to include the caution statements that dapagliflozin should not be used in patients with severe renal impairment and that the use of dapagliflozin in patients with moderate renal impairment should be carefully considered in the Precautions for Indications section of the package insert, and to list patients with moderate renal impairment in the Careful Administration section. PMDA confirmed that appropriate actions have been taken [see “(6) Draft risk management plan” for post-marketing investigations].

2) Patients with hepatic impairment

PMDA concluded that it is necessary to continue to collect information on the safety in patients with hepatic impairment via post-marketing surveillance because dapagliflozin has not been studied in patients with severe hepatic impairment in Japanese clinical studies. In addition, careful administration of dapagliflozin should be recommended in patients with severe hepatic impairment, because the exposure (AUC) increases by approximately 60% in such patients compared with healthy subjects.

The above conclusion of PMDA was supported by the expert advisors.

Based on the above, PMDA instructed the applicant to list patients with severe hepatic impairment in the Careful Administration section, and confirmed that appropriate actions have been taken [see “(6) Draft risk management plan” for post-marketing investigations].

3) Elderly patients

PMDA concluded that, although there is no major problem with the safety in elderly patients on the premise that appropriate caution statements are provided, it is necessary to continue to collect information on the safety in elderly patients via post-marketing surveillance.

The above conclusion of PMDA was supported by the expert advisors [see “(6) Draft risk management plan” for post-marketing investigations].

(6) Draft risk management plan

Taking account of the “4.(iii).B.(7) post-marketing investigations” of the Review Report (1) and the comments from the expert advisors at the Expert Discussion, PMDA concluded that the following points should be additionally evaluated through the risk management plan.

- The impact of concomitant drugs on the safety depending on their doses and types
- Polyuria/pollakiuria
- The impact of weight decreased on the safety
- The impact of increased ketone bodies
- The safety and efficacy in patients with renal or hepatic impairment
- A focused survey on adverse events in all elderly patients treated with dapagliflozin

PMDA instructed the applicant to take actions on the above points, and the applicant presented the risk management plan (Tables 37, 38) and specified drug use-results survey plan (draft) (Tables 39, 40) as a response. PMDA confirmed that the contents of these plans were appropriate.

Table 37. Safety and efficacy specifications of the draft risk management plan

Safety specifications		
Important identified risks	Important potential risks	Important missing information
<ul style="list-style-type: none">• Hypoglycaemia• Genital infection• Urinary tract infection• Polyuria/pollakiuria• Events related to volume depletion	<ul style="list-style-type: none">• The impact of weight decreased on the safety• The impact of increased ketone bodies• Renal disorder• Liver disorder• Fracture• Malignant tumor	<ul style="list-style-type: none">• Safety in elderly patients during treatment• Safety in patients with renal impairment during treatment• Safety in patients with hepatic impairment during treatment• Safety in patients with cardiac failure during treatment• Safety during concomitant therapy with dapagliflozin and insulin products
Efficacy specifications		
<ul style="list-style-type: none">• Long-term efficacy in routine clinical settings• Reduction of incidence of major cardiovascular events• Efficacy of concomitant therapy with dapagliflozin and insulin products		

Table 38. Summary of additional pharmacovigilance activities and risk minimization activities in the risk management plan

Additional pharmacovigilance activities	Additional risk minimization activities
<ul style="list-style-type: none"> • Early post-marketing phase vigilance • Specified drug use-results survey in elderly patients • Specified drug use-results survey of long-term use • Global study on reduction of incidence of major cardiovascular events (DECLARE Study) (footnote 167) • Post-marketing clinical study^{a)} 	<ul style="list-style-type: none"> • Early post-marketing phase vigilance • Preparation and provision of materials for patients • Preparation and provision of materials for health-care providers

a) After dapagliflozin is approved, a clinical study evaluating concomitant therapy with dapagliflozin and insulin products will be conducted.

Table 39. Outline of specified drug use-results survey of long-term use (draft)

Objective	Confirm the long-term safety and efficacy of dapagliflozin for 3 years
Survey method	Central registry system
Patients population	Patients with type 2 diabetes mellitus
Observation period	3 years
Target sample size	6000 patients (3000 patients to be evaluated in 3 years)
Priority investigation items	Patient characteristics, exposure to dapagliflozin, concomitant drugs, efficacy assessment (e.g., HbA1c), safety assessment (e.g., effect on cardiovascular system, malignant tumor, other adverse events)

Table 40. Outline of specified drug use-results survey in elderly patients (draft)

Objective	Confirm the safety of dapagliflozin in elderly patients
Survey method	Central registry system
Patients population	Patients with type 2 diabetes mellitus aged ≥ 65 years
Observation period	1 year
Target sample size	All patients treated with dapagliflozin during the first 3 months after the market launch
Priority investigation items	Patient characteristics, exposure to dapagliflozin, concomitant drugs, safety assessment (e.g., adverse events related to volume depletion, urinary tract infections, other adverse events)

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 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 took place in accordance with the provisions of the Pharmaceutical Affairs Act for the data submitted in the new drug application (5.3.5.1.1, 5.3.5.1.2, 5.3.5.2.1). As a result, protocol deviations (enrollment of subjects who did not meet the inclusion criteria, non-compliance with the rules for pregnancy testing, non-compliance with the rules for treatment assignment) were found at some trial sites. In addition, some of the above deviations were not found to be appropriately detected by the sponsor's monitors. Although these findings requiring improvement were noted, PMDA concluded that the clinical studies as a whole were performed in compliance with GCP and there should be no problem with conducting a regulatory review based on the submitted application documents.

IV. Overall Evaluation

As a result of the above review, PMDA has concluded that dapagliflozin may be approved for the following indication and dosage and administration. The re-examination period is 8 years. Neither the drug substance nor the drug product is classified as a poisonous drug or a powerful drug, and the product is not classified as a biological product or a specified biological product.

[Indication]	Type 2 diabetes mellitus
[Dosage and administration]	The usual adult dosage is 5 mg of dapagliflozin administered orally once daily. The dose may be increased to 10 mg once daily for the patient with an inadequate response to the 5 mg dose if the patient is followed up carefully.