Report on the Deliberation Results

September 2, 2013

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

[Brand name]	Kadcyla Intravenous Infusion 100 mg Kadcyla Intravenous Infusion 160 mg
[Non-proprietary name]	Trastuzumab Emtansine (Genetical Recombination) (JAN*)
[Applicant]	Chugai Pharmaceutical Co., Ltd.
[Date of application]	January 29, 2013

[Results of deliberation]
In the meeting held on August 26, 2013, the Second Committee on New Drugs concluded that the product may be approved and that this result should be presented to the Pharmaceutical Affairs Department of the Pharmaceutical Affairs and Food Sanitation Council.

The re-examination period is 8 years, the drug substance and the drug product are both classified as powerful drugs, and the product is classified as a biological product.

**Japanese Accepted Name (modified INN)*

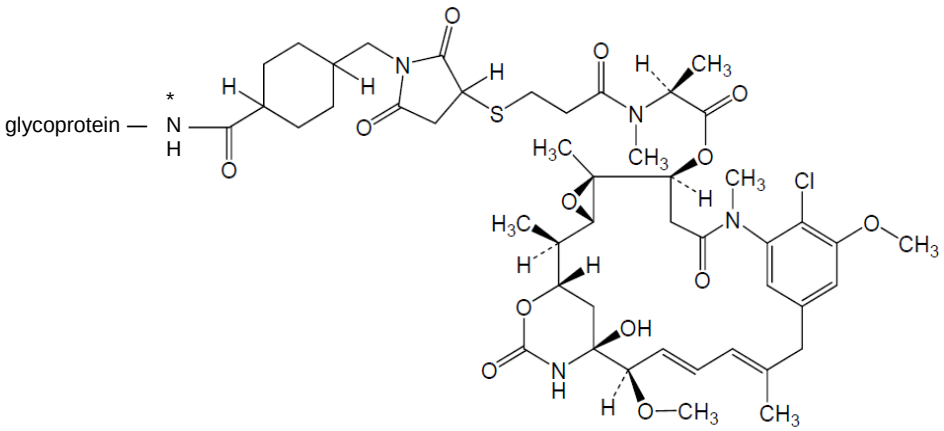
Review Report

August 15, 2013
Pharmaceuticals and Medical Devices Agency

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

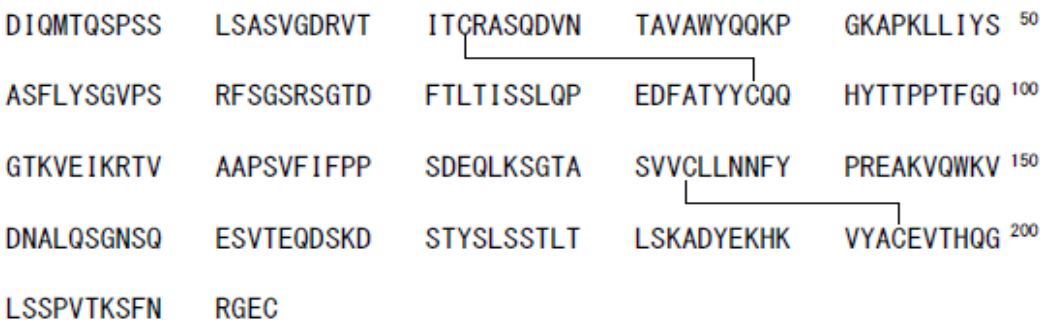
[Brand name]	Kadcyla Intravenous Infusion 100 mg Kadcyla Intravenous Infusion 160 mg
[Non-proprietary name]	Trastuzumab Emtansine (Genetical Recombination)
[Applicant]	Chugai Pharmaceutical Co., Ltd.
[Date of application]	January 29, 2013
[Dosage form/Strength]	Lyophilized powder for solution for intravenous infusion: Each vial contains 106 mg or 171 mg of Trastuzumab Emtansine (Genetical Recombination).
[Application classification]	Prescription drug (1) Drug with a new active ingredient

[Chemical structure]



*: An amino acid residue of trastuzumab

The amino acid sequence of trastuzumab and disulfide bonds within the sequence is as follows.



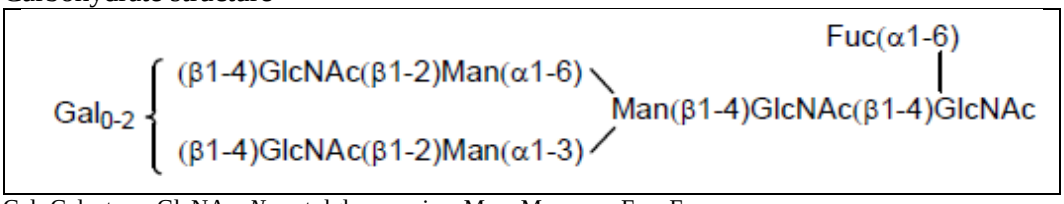
Light chain

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

Heavy chain

Sites that can bind to the drug: D1 in light chain, E1 in heavy chain, and K in light and heavy chains
Glycosylation site: N300 in heavy chain
Partial deficiency: K450 in heavy chain
Disulfide bonds: C214 in light chain – C223 in heavy chain, C229 in heavy chain – C229 in heavy chain, C232 in heavy chain – C232 in heavy chain

Carbohydrate structure



Gal: Galactose, GlcNAc: *N*-acetylglucosamine, Man: Mannose, Fuc: Fucose

Molecular formula: Emtansine: $C_{47}H_{62}ClN_4O_{13}S$
Trastuzumab (Genetical Recombination): $C_{6460}H_{9972}N_{1724}O_{2014}S_{44}$

Molecular weight: Approx. 151,000

Chemical name: Trastuzumab Emtansine is an antibody-drug conjugate (molecular weight: ca. 151,000) consisting of Emtansine attached mainly to the ε-amino group of an average of 3.5 Lys residues of Trastuzumab (Genetical Recombination).
Emtansine (4-({3-[(3-{[(1*S*)-2-{[(1*S*,2*R*,3*S*,5*S*,6*S*,16*E*,18*E*,20*R*,21*S*)-11-chloro-21-hydroxy-12,20-dimethoxy-2,5,9,16-tetramethyl-8,23-dioxo-4,24-dioxa-9,22-diazatetracyclo[19.3.1.$1^{10,14}$.$0^{3,5}$]hexacosa-10,12,14(26),16,18-pentaen-6-yl]oxy}-1-methyl-2-oxoethyl]methylamino}-3-oxopropyl)sulfanyl]-2,5-dioxopyrrolidin-1-yl}methyl)cyclohexylcarbonyl [$C_{47}H_{62}ClN_4O_{13}S$; molecular weight: 958.53]) is a maytansinoid DM1 conjugated to a 4-[(2,5-dioxopyrrolidin-1-yl)methyl] cyclohexylcarbonyl linker.

[Items warranting special mention]	Priority review (Notification No. 0220-2 from the Evaluation and Licensing Division, Pharmaceutical and Food Safety Bureau, MHLW, dated February 20, 2013)
[Reviewing office]	Office of New Drug V

Review Results

August 15, 2013

[Brand name]	Kadcyla Intravenous Infusion 100 mg Kadcyla Intravenous Infusion 160 mg
[Non-proprietary name]	Trastuzumab Emtansine (Genetical Recombination)
[Applicant]	Chugai Pharmaceutical Co., Ltd.
[Date of application]	January 29, 2013

[Results of review]
Based on the submitted data, it is concluded that the efficacy of the product in patients with HER2-positive inoperable or recurrent breast cancer has been demonstrated and its safety is acceptable in view of its observed benefits. The occurrence of thrombocytopenia, hepatotoxicity, etc., needs to be further investigated via post-marketing surveillance.

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

[Indication]	HER2-positive inoperable or recurrent breast cancer
[Dosage and administration]	The usual adult dosage is 3.6 mg/kg (body weight) of Trastuzumab Emtansine (Genetical Recombination) given as an intravenous infusion every 3 weeks.

Review Report (1)

July 8, 2013

I. Product Submitted for Registration

[Brand name]	Kadcyla Intravenous Infusion 100 mg Kadcyla Intravenous Infusion 160 mg
[Non-proprietary name]	Trastuzumab Emtansine (Genetical Recombination)
[Applicant]	Chugai Pharmaceutical Co., Ltd.
[Date of application]	January 29, 2013
[Dosage form/Strength]	Lyophilized powder for solution for intravenous infusion: Each vial contains 100 mg or 160 mg of Trastuzumab Emtansine (Genetical Recombination).
[Proposed indication]	HER2-positive inoperable or recurrent breast cancer
[Proposed dosage and administration]	The usual adult dosage is 3.6 mg/kg (body weight) of Trastuzumab Emtansine given as an intravenous infusion every 3 weeks.

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

A summary of the submitted data and the outline of the review by the Pharmaceuticals and Medical Devices Agency (PMDA) are as shown below.

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

1.(1) Drug overview

Trastuzumab Emtansine (Genetical Recombination) (hereinafter referred to as trastuzumab emtansine) is an antibody-drug conjugate developed by Genentech Inc. (the US), the company that discovered trastuzumab (genetical recombination) (trastuzumab), which is a humanized monoclonal antibody directed against human epidermal growth factor receptor type 2 (HER2). The conjugate is comprised of trastuzumab, which is bound to *N*2'-deacetyl-*N*2'-(3-mercapto-1-oxopropyl)-maytansine (DM1), a tubulin polymerization inhibitor synthesized from ansamitocin P3 of microbiological origin, via 4-(*N*-maleimidomethyl) cyclohexane-1-carboxylate (MCC) as a linker.

Trastuzumab emtansine, upon binding to HER2, induces antibody-dependent cell-mediated cytotoxicity, etc., as is the case with trastuzumab and then DM1, thus incorporated into cells, inhibits tumor growth by inducing cell cycle arrest and apoptosis.

1.(2) Development history, etc.

In foreign countries, a phase I study (Study TDM3569g) was initiated by Genentech, Inc. (US) in April 2006, involving patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including trastuzumab. Also, a phase II study (Study TDM4258g) was started in [redacted], [redacted] involving patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including HER2-targeted therapy, and another phase II study (Study TDM4374g) was initiated in [redacted], [redacted] involving patients with HER2-positive metastatic breast cancer who had previously been treated with chemotherapy with anthracyclines and taxane antineoplastic drugs, capecitabine, trastuzumab, and lapatinib tosilate hydrate. Subsequently, a phase III study (Study TDM4370g/Study BO21977, [EMILIA study]) was commenced in February 2009, involving patients with HER2-positive metastatic or recurrent breast cancer who had previously been treated with chemotherapy with taxane antineoplastic

drugs and trastuzumab.

In August 2012, a marketing application for trastuzumab emtansine including the pivotal data from the EMILIA study has been filed by Genentech in the US and by F. Hoffmann-La Roche (Roche) in the EU. In the US, trastuzumab emtansine was approved in February 2013 for the following indication: "KADCYLA, is a HER2-targeted antibody and microtubule inhibitor conjugate indicated, as a single agent, for the treatment of patients with HER2-positive, metastatic breast cancer who previously received trastuzumab and a taxane, separately or in combination. Patients should have either: received prior therapy for metastatic disease, or developed disease recurrence during or within six months of completing adjuvant therapy." In the EU, trastuzumab emtansine is currently under review.

As of May 2013, trastuzumab emtansine has been approved in 3 countries with the indication for the treatment of breast cancer.

In Japan, a phase I study (Study JO22591) and a phase II study (Study JO22997) were initiated by the applicant in [redacted], [redacted] and in [redacted], [redacted], respectively, involving patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including trastuzumab.

In January 2013, a marketing application for trastuzumab emtansine including the pivotal data from the EMILIA study has been filed.

2. Data relating to quality

2.A. Summary of the submitted data

2.A.(1) Drug substance

The drug substance trastuzumab emtansine (genetical recombination) is an antibody-drug conjugate consisting of *N*2'-deacetyl-*N*2'-(3-mercapto-1-oxopropyl)-maytansine (DM1) attached to trastuzumab (genetical recombination) (trastuzumab), a monoclonal antibody against human epidermal growth factor receptor type 2 (HER2), via 4-(*N*-maleimidomethyl) cyclohexane-1-carboxylate (MCC) as a linker. [redacted].

2.A.(1).1) Trastuzumab

The manufacturing process of trastuzumab is identical to that of the drug substance of approved drugs "Herceptin Intravenous Infusion 60" and "Herceptin Intravenous Infusion 150." [redacted]. No new data on the manufacturing and control of trastuzumab were submitted in the present application.

2.A.(1).2) DM1

2.A.(1).2).i) Characterization

DM1 is white powder and its properties including description, solubility, hygroscopicity, melting point, acid dissociation constant, partition coefficient, optical rotation, and crystalline polymorphism have been determined.

The chemical structure of DM1 has been elucidated by ultraviolet spectrophotometry (UV), infrared spectrophotometry (IR), elemental analysis, mass spectrometry, nuclear magnetic resonance spectroscopy (^{1}H-, ^{13}C-, [redacted]), and single-crystal X-ray crystallography.

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

[redacted].

DM1 was developed using quality-by-design (QbD) approach whereby mainly the following investigations were performed.

- [redacted]
- Identification of the criticality of manufacturing process parameters and process control, based on risk assessment
- Development of a control strategy

[redacted].

2.A.(1).2).iii) Control

[redacted].

During the review process, optical rotation was included as a specification.

2.A.(1).3) Trastuzumab emtansine (genetical recombination)

2.A.(1).3).i) Manufacturing process

[redacted].

The manufacturing process was developed according to the QbD approach [see "2.A.(4) Quality by Design (QbD)].

[redacted].

The manufacturing process is evaluated at production scale.

2.A.(1).3).ii) Manufacturing process development (comparability)

The major changes made in the manufacturing process in the course of the drug substance development are as follows:

- From manufacturing process A to process B:

  [redacted]

- From manufacturing process B to process C:

[redacted]

- From manufacturing process C to process D:
  [redacted]

- [redacted]

The comparability of the quality attributes of the drug substance was evaluated before and after these changes in the manufacturing process.

2.A.(1).3).iii) Characterization

(a) Structure and composition

[redacted]

- [redacted].

Mass spectrometry

- [redacted].

Distribution of the quantity of drug conjugated forms

- [redacted].

Peptide mapping

- [redacted].

- [redacted].

Free maytansinoids

- [redacted].

Higher-order structure

- [redacted].
- [redacted].

Carbohydrate structure

- [redacted].

(b) Physicochemical properties

Electrophoresis

- [redacted].
- [redacted].

Liquid chromatography

- [redacted].
- Size exclusion chromatography (SE-HPLC) showed peaks of high and low molecular weight forms, in addition to the main peak.

(c) Biological properties

- [redacted].
- Surface plasmon resonance analysis showed that HER2 binding constant, dissociation rate constant, and binding rate constant of the drug substance were equivalent to those of trastuzumab.

- [redacted].
- [redacted].
- [redacted].
- [redacted].
- [redacted].
- [redacted].
- [redacted].
- Flow cytometry of SK-BR-3 and BT-474 cell lines derived from HER2-positive human breast cancer showed that trastuzumab arrested the cell cycle at the G0/G1 phase, whereas the drug substance and a high concentration of DM1 arrested the cell cycle at the G2/M phase.
- The drug substance showed a dose-dependent apoptosis-inducing activity (caspase 3/7 activity) in SK-BR-3 and BT-474 cell lines. In contrast, trastuzumab did not induce apoptosis in these cell lines.
- [redacted]. In HER2 low-expressing cell lines, cytocidal activity was observed only at high concentrations.
- [redacted].

2.A.(1).3).iv) Process-related impurities

[redacted]

[redacted].

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

[redacted].

2.A.(1).3).vi) Stability of drug substance

The main stability studies for the drug substance are as shown in the table below.

Outline of main stability studies for drug substance

	Manufacturing process	Number of batches	Storage conditions	Study period	Storage configuration
Long-term testing	Manufacturing process D	1	-20°C	36 months	[redacted] tank
		1		[redacted] months*	
	Commercial manufacturing process	3		[redacted] months*	
		3		[redacted] months*	
Accelerated testing	Manufacturing process D	2	5 ± 3°C	3 months	
	Commercial manufacturing process	3		3 months	
		3		6 months	
Stress testing	Manufacturing process D	1	25°C/60% RH	1 month	Glass vials
		3	40°C/75% RH		
	Commercial manufacturing process	6			

*: Ongoing

The long-term testing showed no significant changes in the quality attributes throughout the study period.

The accelerated testing showed increased levels of free maytansinoids.

[redacted].

[redacted].

2.A.(2) Drug product

2.A.(2).1) Drug product, formulation, and formulation development

The drug product is an injectable formulation containing 106 mg of the drug substance in one 15-mL glass vial or 171 mg of the drug substance in one 20-mL glass vial. The drug product contains purified sucrose, succinic acid, and polysorbate 20 as excipients. The drug product is supplied with Water for Injection (JP grade, 5 or 8 mL) filled in glass ampules or polyethylene containers. Each vial is overfilled to ensure that the labeled amount (100 or 160 mg) of the drug substance

can be withdrawn after reconstitution.

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

[redacted].

The manufacturing process was developed using QbD approach [see "2.A.(4) Quality by Design [QbD]"].

[redacted].

The manufacturing process was evaluated on a commercial scale.

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

[redacted].

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

[redacted].

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

The main stability studies for the drug product are as shown in the table below.

Outline of the stability studies for drug product

	Formulation	Number of batches	Storage conditions	Study period	Storage configuration
Long-term testing	100 mg	3	5 ± 3°C	[redacted] months*1	Glass vials
	160 mg	4		[redacted] months*1	
		1		36 months	
Accelerated testing	100 mg	3	25°C/60% RH	6 months	
	160 mg	5			
Stress testing	100 mg	3	50°C/75% RH	6 months	
	160 mg	4			
Photostability testing	100 mg	1	≥1.20 million lux·h and ≥200 W·h/m^2 as integrated near ultraviolet energy		Glass vials and glass vials light-shielded with aluminum foil
	160 mg	1			
[redacted]	100 mg	1	[redacted]		Glass vials
	160 mg	1			

*1: [redacted]

The long-term testing showed no significant changes in quality attributes throughout the study period.

The accelerated testing showed no significant changes in quality attributes of the 100-mg formulation throughout the study period but showed increased levels of free maytansinoids in the 160-mg formulation.

[redacted].

The photostability testing showed no significant changes in the quality attributes under the conditions investigated.

[redacted].

[redacted]. The long-term testing will be continued up to 48 months.

2.A.(3) Reference materials

[redacted].

[redacted].

2.A.(4) Quality by Design (QbD)

QbD approach was employed for the development of the drug substance and the drug product. The strategies for quality control were developed based mainly on the following investigations.

- [redacted].
- [redacted].
- [redacted].
- [redacted].

- [redacted].

2.B. Outline of the review by PMDA

PMDA reviewed the submitted data including the following major issues evaluated. As a result, it was concluded that, although the appropriateness of the shelf life of the 100-mg formulation should be further investigated, other quality aspects of the drug substance and the drug product are controlled in an appropriate manner.

2.B.(1) Starting materials for DM1

DM1 is to be manufactured at [redacted] manufacturing sites, and the method for the controlling starting materials varies depending on the sites. PMDA therefore asked the applicant to explain the background to the setting of different controlling methods and the appropriateness thereof.

The applicant responded as follows:
The manufacturing process was developed independently at each manufacturing site, which resulted in minor differences in the manufacturing process, albeit considerable similarity. There are also some differences in the impurity profiles of the starting materials used. [redacted]. However, since the control strategy developed by each manufacturing site is appropriately in place, and is capable of consistently manufacturing DM1 with appropriate quality even from the differently controlled starting materials.

PMDA accepted the applicant's above response by taking account of the controlling method for the starting materials, manufacturing process, the extent of contribution of the controlling method for the starting materials to the control strategy, the results of batch analysis, etc. at each manufacturing site.

2.B.(2) Shelf life of drug substance and drug product

The applicant had proposed a shelf life of 36 months for both drug substance and drug product, based on the data of the following long-term testing and examination thereof. However, the shelf life of the drug substance and the drug product should be based on long-term stability data on at least 3 batches. Therefore, PMDA requested the applicant to re-determine the shelf life based on the most updated long-term stability data.

- Results of long-term testing on 1 batch of the drug substance up to 36 months, long-term testing on 3 batches of the drug substance up to [redacted] months, etc.
- [redacted].

The applicant explained as follows:
On the basis of the additionally submitted data including the results of long-term testing on 3 batches of the drug substance manufactured by the commercial-scale manufacturing process (up to 36 months), results of long-term testing on 3 batches of 100-mg formulation (up to 30 months), and results of a long-term testing on 3 batches of 160-mg formulation (up to 36 months), neither the drug substance nor the drug product showed significant changes in any test parameters

throughout the study period. Therefore, the applicant determined the shelf life of 36 months for the drug substance when stored at -20°C, and the shelf life of 30 months and 36 months for the 100-mg and 160-mg formulations, respectively, when stored at 5 ± 3°C.

[redacted].

PMDA considers as follows:
The proposed shelf lives for the drug substance and the 160-mg formulation are acceptable. For the 100-mg formulation, however, the submitted data are partly insufficient for setting the shelf life as proposed. The appropriateness of the proposed shelf life should be further investigated.

2.B.(3) Identification of clinical quality attributes

[redacted].

PMDA considers as follows:
The criticality of the quality attributes should not change depending on control elements. Instead, quality attributes should be evaluated based on the magnitude of their potential impact on the efficacy and safety. Therefore, the applicant's above explanation is inappropriate. [redacted].

3. Non-clinical data

3.(i) Summary of pharmacology studies

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

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

3.(i).A.(1).1) Tumor growth inhibition

***in vitro*:**

Tumor growth-inhibitory effect on HER2-positive breast cancer cell lines (Report [redacted]-0825)

The tumor growth-inhibitory effect of Trastuzumab Emtansine (Genetical Recombination) (hereinafter referred to as trastuzumab emtansine) on human breast cancer-derived (a) SK-BR-3, (b) BT-474, (c) KPL-4, (d) HCC1954, (e) BT-474EEI, (f) MCF7, and (g) MDA-MB-468 cell lines was investigated. Cell lines (a) to (e) above were used as those expressing high levels of human epidermal growth factor receptor type 2 (HER2) (positive, [a] to [d] 3+ and [e] 2+ by flow cytometry [FCM]), (f) as the cell line expressing low levels of HER2 (normal level, 0 by FCM), and (g) as the cell line not expressing HER2 (negative by FCM). Trastuzumab (genetical recombination) (trastuzumab) was used as the control.

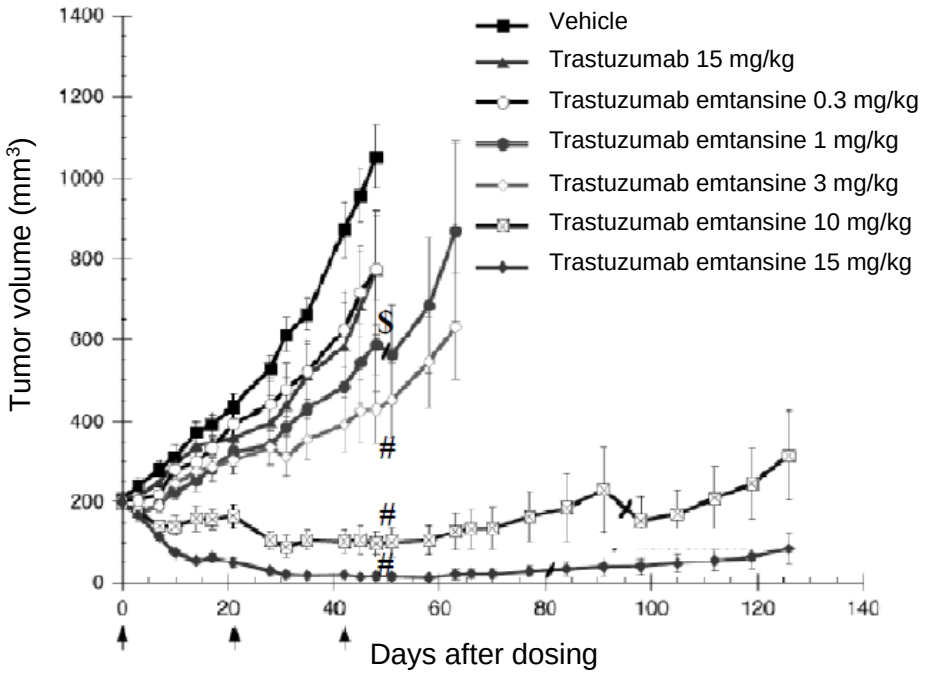
Results showed that trastuzumab emtansine inhibited the growth of trastuzumab-resistant cell lines ([c], [d], [e]) and inhibited the growth of trastuzumab-sensitive cell lines ([a], [b]) more potently than trastuzumab did. In contrast, trastuzumab emtansine did not inhibit the growth of HER2 low-expressing or non-expressing cell lines ([f], [g]).

***in vivo*:**

Tumor growth-inhibitory effect in mice transplanted with HER2-postive breast cancer cell lines (Report [redacted]-0962-1459, [redacted]-1629)

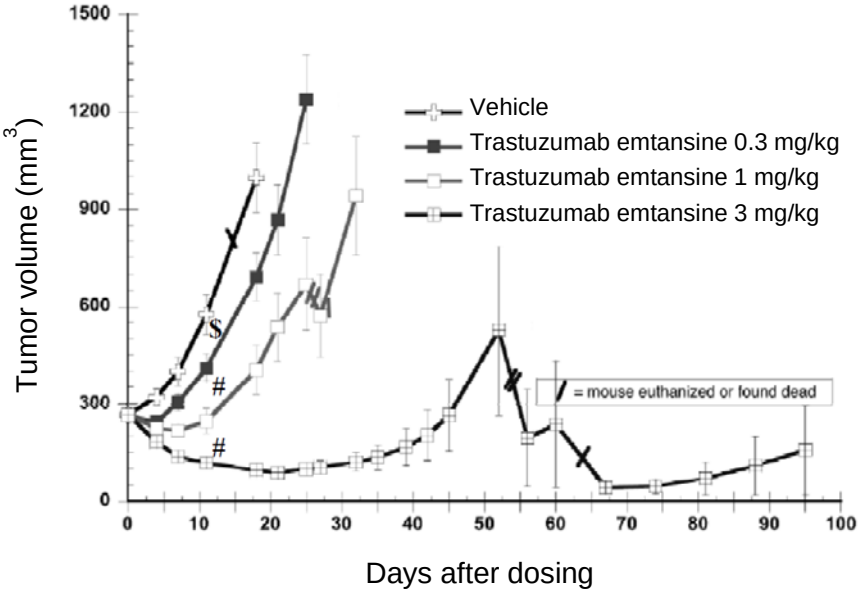
Beige mice with reduced natural killer activity were mated with athymic mice (nude mice) to generate a mouse model (beige-nude mouse). The beige-nude mice were orthotopically

transplanted with the BT-474EEI cell line (trastuzumab-resistant, estrogen-independent) and subjected to the test for the tumor growth-inhibitory effect of trastuzumab emtansine. Starting on Post-transplantation day 11 when the transplanted tumor reached approximately 200 mm^3 in volume, trastuzumab emtansine (0.3, 1, 3, 10, 15 mg/kg) was administered intravenously once every 3 weeks for 3 doses in total, and tumor volume was calculated (the figure below). Control animals received vehicle or trastuzumab (15 mg/kg) intravenously once every 3 weeks for 3 doses in total.



Tumor growth-inhibitory effect of trastuzumab emtansine (BT-474EEI cell line)
Mean ± standard error (SE), n = 10 (number of animals at the initiation of dosing)
Each arrow indicates the day of administration of trastuzumab emtansine, trastuzumab, or vehicle, each slash on the graph indicates that an animal was euthanized because the tumor volume exceeded 1000 mm^3 or for any other reason.
$: $P < 0.05$ (Tukey test) against the vehicle group
#: $P < 0.001$ (Tukey test) against the vehicle group (The statistical analysis was performed on the tumor volume at the last measurement day when all animals were alive [48 days after the initiation of dosing].)

Beige mice were mated with severe combined immunodeficiency (SCID) mice to generate a mouse model (SCID-beige mouse). The SCID-beige mice were orthotopically transplanted with the KPL-4 cell line (trastuzumab-resistant) and subjected to the test for the tumor growth-inhibitory effect of trastuzumab emtansine. On Post-transplantation day 11 when the mean volume of the transplanted tumor reached 265 (197-369) mm^3, trastuzumab emtansine (0.3, 1, 3 mg/kg) was administered intravenously in a single dose, and tumor volume was calculated (the figure below). Control animals received a single intravenous dose of the vehicle.



Tumor growth-inhibitory effect of trastuzumab emtansine (KPL-4 cell line)

Mean ± SE, n = 8 (number of animals at the initiation of dosing)
Each slash on the graph indicates that an animal was euthanized because the tumor volume exceeded 1000 mm^3 or for any other reason.
$: $P < 0.05$ (Tukey test) against the vehicle control group
#: $P < 0.001$ (Tukey test) against the vehicle control group (The statistical analysis was performed on the tumor volume at the last measurement day when all animals were alive [11 days after the initiation of dosing].)

The applicant explained that the results of the *in vitro* and *in vivo* studies demonstrated the tumor growth-inhibitory activity of trastuzumab emtansine against HER2-positive human breast cancer cell lines which are resistant to trastuzumab.

3.(i).A.(1).2) Mechanism of action

i) Binding characteristics to HER2 (Report [redacted]-0010, [redacted]-1111)

[redacted]. The dissociation constant (K_D) of trastuzumab emtansine and trastuzumab was 1.08 ± 0.19 and 1.01 ± 0.18 nmol/L, respectively, showing a similar binding activity.

[redacted].

ii) Binding activity with FcγR and antibody-dependent cell-mediated cytotoxicity (Report [redacted]-0046)

Soluble forms of main human Fcγ receptors (FcγRs) were prepared and their binding activity with trastuzumab emtansine was investigated by enzyme-linked immunosorbent assay (ELISA). Against the high-affinity receptor FcγRIa, the 50% effective concentration (EC_{50}) of trastuzumab emtansine and trastuzumab, calculated from the binding curve, was 18.9 ± 3.46 and 24.0 ± 0.96 ng/mL, respectively, showing a similar binding activity. Against the low-affinity receptors FcγRIIa, FcγRIIb, and FcγRIIIa, the binding activity of trastuzumab emtansine was slightly greater (approximately 2- to 6-fold) compared with that of trastuzumab. The applicant explained that these changes were possibly due to the differences in the carbohydrate structure.

Using the fluorescence-labeled HER2-positive BT-474 cell line as the target cells (T) and peripheral mononuclear cells collected from healthy adult subjects as effector cells (E), a mixture of these cells (E:T = 25:1) was incubated in the presence of trastuzumab emtansine or trastuzumab for 3 hours to investigate antibody-dependent cell-mediated cytotoxic (ADCC) activity. Trastuzumab emtansine showed a slightly more potent ADCC activity compared with trastuzumab. However, trastuzumab emtansine and trastuzumab derived from the same batch of the drug substance showed similar ADCC activity to each other [see "2.A.(1).3).iii).(c) Biological properties], from which the applicant explained that the observed difference may be due to the difference of batches of the drug substance and that the binding of trastuzumab to DM1 via a linker does not affect ADCC activity.

iii) Effect of trastuzumab emtansine on cell cycle, apoptosis, PI3K/AKT signal transduction, and HER2 ECD shedding (Report [redacted]-0825)

The following data were submitted.

- Using SK-BR-3, BT-474, and KPL-4 cell lines, the effect of trastuzumab emtansine on cell cycle was investigated in terms of the number of cells in the G2/M phase after the cells were treated with the drug for 24 hours (KPL-4 cell line) or for 48 hours (SK-BR-3, BT-474 cell lines). As a result, the number of cells in the G2/M phase did not change after treatment with trastuzumab (control), but increased after treatment with trastuzumab emtansine.

- Using SK-BR-3, BT-474, and KPL-4 cell lines, the apoptosis-inducing activity of trastuzumab emtansine was investigated in terms of activation of caspase 3/7 after 48-hour treatment. As a result, apoptosis was not induced by treatment with trastuzumab (control) but induced by treatment with trastuzumab emtansine.

- Using the KPL-4 cell line, the effect of trastuzumab emtansine on cell cycle and apoptosis-inducing activity were investigated in terms of histone H3 phosphorylation (marker of the G2/M phase) and decrease in XIAP and cleavage of PARP (apoptosis markers). Results showed that trastuzumab emtansine arrested cell cycle at the G2/M phase and induced apoptosis.

- Using the BT-474-M1 cell line, established by passaging the BT-474 cell line in nude mice, and the KPL-4 cell line, the effect of trastuzumab emtansine on AKT phosphorylation was investigated. Treatment with trastuzumab emtansine inhibited AKT phosphorylation.

- Using the BT-474-M1 cell line, the effect of trastuzumab emtansine on HER2 ECD shedding was investigated. The IC_{50} of trastuzumab emtansine was 0.18 ± 0.04 μg/mL, showing an inhibitory activity similar to that of trastuzumab used as the control (IC_{50}, 0.30 ± 0.05 μg/mL).

iv) Characterization of trastuzumab emtansine as an antibody-drug conjugate (Reports [redacted]-1508, [redacted]-292ZF-1469, [redacted]-292X-1459)

In order to characterize trastuzumab emtansine as an antibody-drug conjugate consisting of trastuzumab linked to *N*2'-deacetyl-*N*2'-(3-mercapto-1-oxopropyl)-maytansine (DM1) via 4-(*N*-maleimidomethyl) cyclohexane-1-carboxylate (MCC) as a linker, tumor growth-inhibitory effect of a single intravenous administration of (a) trastuzumab emtansine (15 mg/kg), (b) trastuzumab (15 mg/kg), (c) DM1 (184, 454 μg/kg), or (d) combination of trastuzumab (15 mg/kg) and DM1 (184 μg/kg) (trastuzumab/DM1) was investigated using nude mice orthotopically transplanted with mammary tumor tissue (MMTV-HER2 Fo5, 3+ by immunohistochemistry [IHC]) derived from transgenic mice engineered to overexpress HER2 gene in the mammary tissue by a mouse mammary tumor virus (MMTV) promoter. The tumor tissue is resistant to trastuzumab.

Results showed that tumor volume doubling time increased in the trastuzumab emtansine group (27.6 days) compared with the control group (2.8 days). In contrast, the doubling time increased only slightly in the DM1 454 μg/kg group (4.8 days) and did not show any significant increase in the DM1 184 μg/kg group and in the trastuzumab/DM1 group. The applicant explained that concomitant use of DM1 with trastuzumab does not enhance the pharmacological action of DM1, and that the drug substance should be a compound comprising trastuzumab and DM1 as an antibody-drug conjugate in order to exhibit a potent tumor growth-inhibitory effect.

In order to investigate whether or not the tumor growth-inhibitory effect of trastuzumab emtansine is specific to HER2-positive tumors, the tumor growth-inhibitory effect of a single intravenous administration of trastuzumab emtansine (10 mg/kg) and trastuzumab (10 mg/kg) was investigated using beige-nude mice orthotopically transplanted with MMTV-HER2 Fo5. As the control, beige-nude mice orthotopically transplanted with mammary tumor tissue (HER2-negative) derived from transgenic mice engineered to overexpress the *WNT* gene in the mammary tissue by a MMTV promoter were used.

Results showed that the tumor volume doubling time increased only after a single dose of trastuzumab emtansine was administered intravenously to beige-nude mice orthotopically transplanted with HER2-positive MMTV-HER2 Fo5.

v) Investigation of dose and administration method of trastuzumab emtansine (Reports [redacted]-292ZC-1459, [redacted]-292ZD-1459, [redacted]-0962-A-1459)

The relationship between the dosage regimen of trastuzumab emtansine and tumor growth-inhibitory effect was investigated using nude mice orthotopically transplanted with MMTV-HER2 Fo5. Trastuzumab emtansine (1, 3, 10, 15, 30 mg/kg) was administered intravenously once every 3 weeks for a total of 3 doses. The tumor growth-inhibitory effect was observed in the ≥10 mg/kg groups, with the maximum inhibition observed in the 15 and 30 mg/kg groups. Trastuzumab emtansine (3.3, 5, 10 mg/kg) was administered intravenously once every week for a total of 9 doses, and the tumor growth-inhibitory activity was compared with that observed after intravenous dose of 15 mg/kg of trastuzumab emtansine once every 3 weeks for a total of 3 doses. Results showed tumor growth-inhibitory effect even in the group receiving trastuzumab emtansine once weekly at 5 mg/kg, with the most persistent tumor growth-inhibitory effect being observed in the 10 mg/kg once-weekly group.

Trastuzumab emtansine exhibited a potent tumor growth-inhibitory activity also in mice orthotopically transplanted with the BT-474EEI cell line at the same dosage regimen as used in the MMTV-HER2 Fo5 mammary tumor-transplanted model.

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

3.(i).A.(2).1) Effect on central nervous system (Reports [redacted]-0977-1459, [redacted]-0653)

Effect of trastuzumab emtansine (1, 3, 10, 30 mg/kg) on general symptoms and neurological findings was investigated in a 3-month repeat dose study (n = 7/sex/group) and in a 6-month repeat dose study (n = 6/sex/group), both in cynomolgus monkeys [see "3.(iii).A.(2).2) Three-month repeat dose study in cynomolgus monkeys and 3.(iii).A.(2).3) Six-month repeat dose study in cynomolgus monkeys]. Neither general symptoms nor neurological findings were affected by trastuzumab emtansine.

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

i) Effect on hERG current (Report [redacted]-0234)

Effect of DM1 on human ether-a-go-go-related gene (hERG) potassium current was investigated by the patch clamp method using the human embryonic kidney-derived HEK293 cell line introduced with hERG. DM1 inhibited hERG potassium current by 2.5% ± 0.4% at 29.5 μmol/L,

the highest concentration studied (n = 3).

ii) Effect on cardiovascular system (Report [redacted]-1031-1605)

In a single dose study in cynomolgus monkeys (4 females/group), the effect of trastuzumab emtansine (3, 10, 30 mg/kg) on electrocardiogram (ECG) and blood pressure (heart rate, systolic blood pressure, diastolic blood pressure, mean arterial pressure, pulse pressure) was investigated. As a result, a slight increase in blood pressure was observed at 30 mg/kg of trastuzumab emtansine, but the increase was within the range of the baseline level, from which the applicant explained that the observed effect was unlikely to be due to the toxic effect of trastuzumab emtansine. Trastuzumab emtansine had no effect on ECG, blood troponin I or T levels, or creatine kinase.

3.(i).A.(2).3) Effect on respiratory system (Report [redacted]-1031-1605)

In a single dose study in cynomolgus monkeys (4 females/group), the effect of trastuzumab emtansine (3, 10, 30 mg/kg) on respiratory rate and respiratory depth was investigated. Results showed no effect of trastuzumab emtansine.

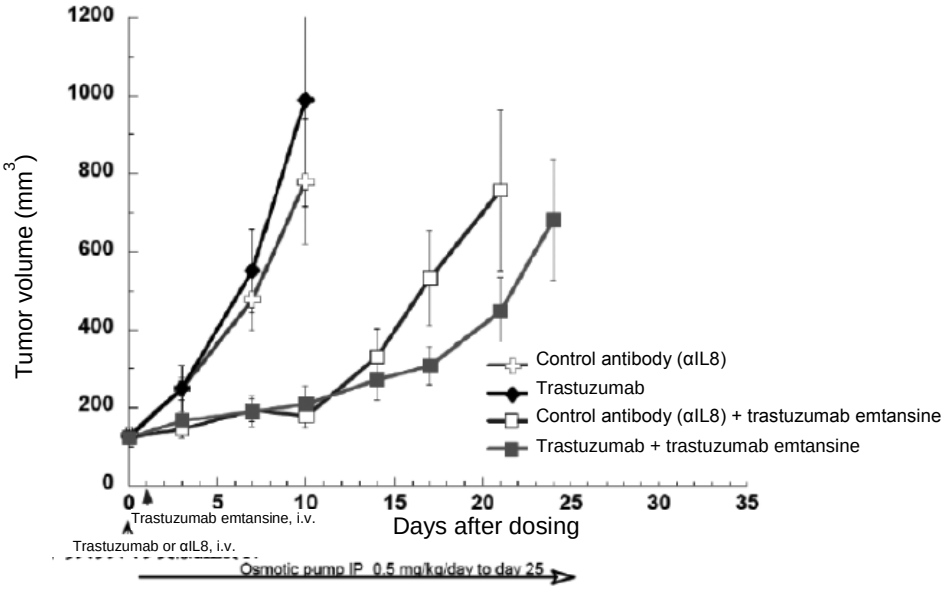
3.(i).A.(2).4) Other (Report [redacted]-1031-1605)

In a single dose study in cynomolgus monkeys (4 females/group), the effect of trastuzumab emtansine (3, 10, 30 mg/kg) on clinical signs, body weight, and food intake was investigated. Results showed no effect of trastuzumab emtansine.

3.(i).A.(3) Pharmacodynamic interaction study

Tumor growth-inhibitory effect of trastuzumab emtansine in the presence of trastuzumab (Report [redacted]-0901-4747)

Since trastuzumab emtansine is intended to be administered to patients with a history of treatment with trastuzumab, whether or not residual trastuzumab affects the tumor growth-inhibitory effect of trastuzumab emtansine was investigated using nude mice orthotopically transplanted with MMTV-HER2 Fo5. Starting on Post-transplantation day 11 when the transplanted tumor reached a mean volume of 122 to 129 mm^3, a single intravenous bolus injection of trastuzumab or negative control antibody (anti-interleukin [IL]-8 antibody) was given at a dose of 15 mg/kg, followed 4 hours later by continuous infusion of the same antibody (0.5 mg/kg/day), which was intraperitoneally infused daily for 25 days using an osmotic pressure pump. To some of these animals, a single intravenous dose of trastuzumab emtansine (10 mg/kg) was administered on the next day of starting trastuzumab or anti-IL-8 antibody dosing, and the tumor volume was calculated (the figure below).



Tumor growth-inhibitory effect of trastuzumab emtansine in the presence of trastuzumab (MMTV-HER2 Fo5)

Mean ± SE, n = 8, Arrows indicate the date or duration of drug administration.

A slightly greater tumor growth-inhibitory effect was observed in the group receiving trastuzumab emtansine in the presence of trastuzumab compared with the group receiving trastuzumab emtansine in the absence of trastuzumab, albeit no statistically significant differences were found (P = 0.1047 [t-test]).

The applicant explained that the above results suggested that trastuzumab did not show any antagonistic effect against the tumor growth-inhibitory effect of trastuzumab emtansine.

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

Based on the submitted data and on the following discussion, PMDA has concluded that trastuzumab emtansine is effective against HER2-positive breast cancer.

Efficacy of trastuzumab emtansine in patients with HER2-positive breast cancer who have a history of treatment with trastuzumab and taxane antineoplastic drugs

Trastuzumab emtansine is intended to be used in breast cancer patients with a history of treatment with trastuzumab, a component of trastuzumab emtansine, and a taxane antineoplastic drug which acts on tubulin as is the case with DM1 [see "4.(iii).B.(4).1) Clinical positioning of trastuzumab emtansine and patients to be treated"]. Therefore, PMDA asked the applicant to explain the efficacy of trastuzumab emtansine in patients with HER2-positive breast cancer that became resistant to trastuzumab and taxane antineoplastic drugs.

The applicant responded as follows:

Trastuzumab emtansine, an antibody-drug conjugate comprising trastuzumab linked to DM1 via a linker, induces antibody-dependent cell-mediated cytotoxicity etc. by binding to HER2, as is the case with trastuzumab and then DM1, thus incorporated into cells, inhibits tumor growth by inducing cell cycle arrest and apoptosis.

DM1 acts on tubulin as is the case with taxane antineoplastic drugs. Whereas trastuzumab emtansine and vinca alkaloid antineoplastic drugs are tubulin destabilizers which inhibit the

stabilization of tubulin by inhibiting the polymerization of tubulin, taxane antineoplastic drugs are tubulin stabilizers which stabilize tubulin by inhibiting the depolymerization of tubulin. Thus, these 2 groups of drugs act by different mechanisms at the molecular level by binding to different sites (*Cancer Treat Rev.* 2012;38:890-903). It is reported that there are two mechanisms whereby cancer cells become resistant to taxane antineoplastic drugs: (a) overexpression of drug transporters such as P-glycoprotein (P-gp), and (b) amino acid mutation in β tubulin molecules (*Semin Oncol.* 2008;35(suppl 2):S1-14, *Cancer Res.* 2010;70:2528-37). In the case of (a), the tumor growth-inhibitory activity of trastuzumab emtansine is likely to be attenuated as is the case with taxane antineoplastic drugs, whereas in case (b), the activity of trastuzumab emtansine is expected to be intact.

Based on the above pharmacological considerations, trastuzumab emtansine is expected to be effective in patients with HER2-positive breast cancer that has become resistant to trastuzumab or taxane antineoplastic drugs.

PMDA accepted the applicant's explanation.

3.(ii) Summary of pharmacokinetic studies

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

Pharmacokinetics (PK) of trastuzumab emtansine was investigated preclinically in mice, rats, and monkeys.

3.(ii).A.(1) Analytical methods

3.(ii).A.(1).1) Measurement of trastuzumab emtansine

[redacted].

3.(ii).A.(1).2) Measurement of trastuzumab

[redacted].

3.(ii).A.(1).3) Measurement of DM1

Concentrations of DM1 released from trastuzumab emtansine (free DM1) in plasma, urine, and bile of mice, rats, and monkeys were measured by LC-MS/MS.

3.(ii).A.(1).4) Measurement of MCC-DM1 and Lys-MCC-DM1

Concentrations of MCC-DM1 and the metabolite generated by the release of MCC-DM1 together with lysine residue (Lys-MCC-DM1) in rat plasma, urine, and bile were measured by LC-MS/MS.

3.(ii).A.(1).5) Measurement of anti-therapeutic antibody (ATA)

[redacted].

3.(ii).A.(2) Absorption

3.(ii).A.(2).1) Single-dose administration

Trastuzumab emtansine (15 mg/kg) was administered intravenously in a single dose to female nude mice, and plasma concentrations of trastuzumab emtansine, total trastuzumab, and free DM1 were measured. After administration, trastuzumab emtansine was eliminated in a biphasic manner,

with CL and $t_{1/2}$ of 13.0 mL/day/kg and 5.6 days, respectively. V_c was 43.8 mL/kg, which was similar to the plasma volume in mice (48.8 mL/kg) (*Handbook of Essential Pharmacokinetics, Pharmacodynamics and Drug Metabolism for Industrial Scientists.* [Bioneer Life Science 2001, CA, USA]). Similarly to trastuzumab emtansine, free DM1 was eliminated in a biphasic manner. The ratio of mean C_{max} of DM1 to that of trastuzumab emtansine was approximately 1/2500. CL and $t_{1/2}$ of total trastuzumab were 7.18 mL/day/kg and 10.9 days, respectively, which were similar to those observed following a single intravenous administration of trastuzumab (10 mg/kg) to female nude mice (CL 6.33 mL/day/kg, $t_{1/2}$ 9.8 days).

Trastuzumab emtansine (0.3, 3, 15 mg/kg) was administered intravenously in a single dose to female nude mice, and plasma concentrations of trastuzumab emtansine, total trastuzumab, and free DM1 were measured (the table below).

The CL and $t_{1/2}$ values of trastuzumab emtansine and total trastuzumab were generally similar regardless of the dose of trastuzumab emtansine, from which the applicant explained that the PK of trastuzumab emtansine and total trastuzumab in mice is linear within the dose range between 0.3 and 15 mg/kg.

PK parameters following single intravenous administration of trastuzumab emtansine to female nude mice*

Dose of trastuzumab emtansine (mg/kg)	0.3	3	15
PK parameters of trastuzumab emtansine			
C_{max} (μg/mL)	5.29	49.3	267
AUC_{inf} (μg·day/mL)	14.3	140	838
CL (mL/day/kg)	18.5	22.1	18.1
V_c (mL/kg)	50.1	62.9	56.8
V_{ss} (mL/kg)	133	164	142
MRT (day)	7.18	7.42	7.85
$t_{1/2}$ (day)	5.19	5.32	5.61
PK parameters of total trastuzumab			
C_{max} (μg/mL)	5.76	53.8	260
AUC_{inf} (μg·day/mL)	32.6	309	1890
CL (mL/day/kg)	8.13	10.0	8.02
V_c (mL/kg)	46.0	57.6	58.2
V_{ss} (mL/kg)	131	165	149
MRT (day)	16.1	16.5	18.6
$t_{1/2}$ (day)	11.4	11.7	13.1
PK parameters of free DM1			
C_{max} (ng/mL)	–	2.09	12.4
AUC_{all} (ng·day/mL)	–	0.294	16.7

Arithmetic mean, n = 4 animals/time point, *: PK parameters were calculated by a 2-compartment model for trastuzumab emtansine and total trastuzumab, and PK parameters for free DM1, by a model-independent analysis.

DM1 (0.1, 0.4 mg/kg) was administered intravenously in a single dose to female nude mice. As a result, CL of free DM1 was 269 and 206 mL/min/mL in the 0.1 and 0.4 mg/kg groups, respectively, and $t_{1/2}$ was 201 and 204 minutes in the 0.1 and 0.4 mg/kg groups, respectively, showing no differences between the 2 groups. V_c was 11,100 and 7920 mL/kg in the 0.1 and 0.4 mg/kg groups, respectively, showing that both values were higher than the body water content in mice (725 mL/kg) (*Pharm Res.* 1993;10:1093-5). The applicant explained that these results suggested the high tissue distribution of DM1.

In order to investigate the effect of the difference in the ratio of DM1 to antibody (drug-antibody ratio [DAR]) on the PK of trastuzumab emtansine, following single intravenous administration of

two types of trastuzumab emtansine with different DAR at the dose of 2 mg/kg to male rats, plasma concentrations of trastuzumab emtansine, total trastuzumab, and free DM1 were measured (the table below). Test samples used in this study were trastuzumab emtansine (DAR, 3.6) with a similar DAR to that of trastuzumab emtansine formulations (DAR, 3.46-3.54) used in clinical studies and trastuzumab emtansine with the lowest possible DAR to be purified (DAR, 2.7). As a result, no clear differences were observed in PK parameters of trastuzumab emtansine, total trastuzumab, and free DM1 between these groups. Based on these results, the applicant explained that the differences of DAR had little effect on the PK of trastuzumab emtansine.

PK parameters after single intravenous administration of trastuzumab emtansine to male rats*

DAR of trastuzumab emtansine	2.7	3.6
PK parameters of trastuzumab emtansine		
C_{max} (μg/mL)	65.6 ± 18.5	60.9 ± 3.55
CL (mL/day/kg)	10.1 ± 0.952	10.5 ± 0.633
V_c (mL/kg)	28.8 ± 8.05	31.0 ± 1.89
V_{ss} (mL/kg)	73.9 ± 9.13	72.9 ± 3.95
MRT (day)	7.28 ± 0.279	6.96 ± 0.138
$t_{1/2}$ (day)	5.43 ± 0.206	5.18 ± 0.204
PK parameters of total trastuzumab		
C_{max} (μg/mL)	62.8 ± 16.2	51.0 ± 2.70
CL (mL/day/kg)	5.07 ± 0.385	6.45 ± 0.511
V_c (mL/kg)	29.8 ± 7.66	37.0 ± 2.04
V_{ss} (mL/kg)	91.4 ± 13.2	92.7 ± 7.27
MRT (day)	18.0 ± 1.72	14.4 ± 1.09
$t_{1/2}$ (day)	13.1 ± 1.37	10.6 ± 0.963
PK parameters of free DM1		
C_{max} (μg/mL)	0.0142 ± 0.00143	0.0214 ± 0.00549

Arithmetic mean ± standard deviation (SD), n = 4, *: PK parameters were calculated by a 2-compartment model for trastuzumab emtansine and total trastuzumab, and PK parameters for free DM1, by a model-independent analysis.

Following single intravenous administion of trastuzumab emtansine (0.3, 3, 20 mg/kg) or single intravenous administion of trastuzumab emtansine (6, 20, 60 mg/kg) to male and female rats, serum concentrations of trastuzumab emtansine and total trastuzumab and plasma concentration of free DM1 were measured (the tables below). In the 60 mg/kg group, 2 males and 1 female died or were euthanized 5 days after administration, and the remaining animals also were euthanized 6 days after administration because of the aggravation of clinical signs. The applicant explained that PK at 60 mg/kg could not be evaluated appropriately for these reasons.

PK of trastuzumab emtansine and total trastuzumab was linear within the dose range from 0.3 to 20 mg/kg. CL of total trastuzumab was approximately 0.5 times that of trastuzumab emtansine. In both experiments, V_c of trastuzumab emtansine and total trastuzumab was similar to the plasma volume in rats (31.3 mL/kg) (*Handbook of Essential Pharmacokinetics, Pharmacodynamics and Drug Metabolism for Industrial Scientists.* [Bioneer Life Science 2001, CA, USA]), and $t_{1/2}$ was constant regardless of the dose. No clear sex differences were observed in the PK of trastuzumab emtansine and total trastuzumab.

Free DM1 concentration in the 0.3 mg/kg group was below the lower limit of quantitation (0.737 ng/mL) at all time points. In other groups, plasma concentrations of free DM1 peaked immediately after administration, then decreased with time. Within the dose range from 3 to 60 mg/kg, C_{max} of free DM1 was generally dose-proportional, while AUC_{all} tended to increase more than dose-proportionally. The applicant explained that the tendency of more than dose-proportional increase in AUC_{all} was due to the limited number of blood sampling points where free DM1 concentration was quantifiable in low dose groups. In the experiment in which 6 to 60

mg/kg of trastuzumab emtansine was administered, free DM1 levels were higher in females than in males in the 20 and 60 mg groups, whereas the levels were higher in males than in females in the 6 mg/kg group, failing to show any consistent trend. The applicant explained that the observed inconsistency was likely to be due to between-assay variation or to inter-individual variability, but the precise reason was unknown. The applicant also explained that, in the experiment in which 0.3 to 20 mg/kg of trastuzumab emtansine was administered, free DM1 was below the lower limit of quantitation at many time points and there were only a limited number of evaluable animals, precluding the evaluation of sex differences in the exposure level of free DM1.

PK parameters after single intravenous administration of trastuzumab emtansine to male and female rats*[1]

Dose of trastuzumab emtansine (mg/kg)	0.3		3		20	
Sex	Male	Female	Male	Female	Male[*2]	Female
PK parameters of trastuzumab emtansine						
C_{max} (μg/mL)	5.39 ± 0.368	5.55 ± 0.122	57.2 ± 5.22	53.1 ± 4.13	398	449 ± 23.9
AUC_{inf} (μg·day/mL)	15.5 ± 1.80	15.3 ± 0.175	165 ± 12.5	151 ± 29.6	1060	1050 ± 134
CL (mL/day/kg)	22.0 ± 3.04	22.1 ± 0.462	19.1 ± 1.75	21.6 ± 3.59	18.4	20.0 ± 3.04
V_c (mL/kg)	62.9 ± 4.24	61.0 ± 1.67	54.9 ± 5.56	60.0 ± 3.66	49.0	45.9 ± 2.44
V_{ss} (mL/kg)	144 ± 17.7	149 ± 2.87	125 ± 15.0	134 ± 16.0	131	126 ± 13.0
MRT (day)	6.58 ± 0.459	6.74 ± 0.205	6.55 ± 0.241	6.25 ± 0.381	7.12	6.36 ± 0.552
$t_{1/2}$ (day)	5.03 ± 0.274	5.00 ± 0.193	5.04 ± 0.229	4.80 ± 0.414	5.16	4.97 ± 0.474
PK parameters of total trastuzumab						
C_{max} (μg/mL)	5.45 ± 0.334	5.90 ± 1.91	51.9 ± 2.20	54.3 ± 5.35	468	444 ± 22.6
AUC_{inf} (μg·day/mL)	32.1 ± 4.30	31.7 ± 2.54	304 ± 16.7	287 ± 65.8	2280	2000 ± 361
CL (mL/day/kg)	10.7 ± 1.65	10.7 ± 0.866	10.3 ± 0.739	11.5 ± 2.36	8.53	10.6 ± 2.51
V_c (mL/kg)	62.1 ± 3.00	57.4 ± 3.24	60.2 ± 2.79	58.8 ± 4.21	41.7	46.4 ± 2.35
V_{ss} (mL/kg)	155 ± 24.2	151 ± 7.63	140 ± 16.6	138 ± 8.95	118	145 ± 8.51
MRT (day)	14.6 ± 1.87	14.2 ± 1.63	13.6 ± 0.968	12.4 ± 1.93	13.8	14.0 ± 2.04
$t_{1/2}$ (day)	10.6 ± 1.35	10.2 ± 1.17	9.91 ± 0.744	9.00 ± 1.38	10.2	10.5 ± 1.56
PK parameters of free DM1						
C_{max} (ng/mL)	–	–	3.77 ± 0.469	4.20 ± 0.519	43.3	27.5 ± 4.82
AUC_{all} (ng·day/mL)	–	–	0.463 ± 0.0342	0.234 ± 0.207	18.4	10.6 ± 1.45

Arithmetic mean ± SD, n = 4, *1: PK parameters were calculated by a 2-compartment model for trastuzumab emtansine and total trastuzumab, and PK parameters for free DM1, by a model-independent analysis. *2: n =1

PK parameters after single intravenous administration of trastuzumab emtansine to male and female rats*[1]

Dose of trastuzumab emtansine (mg/kg)	6		20		60	
Sex	Male	Female	Male	Female[*2]	Male	Female
PK parameters of trastuzumab emtansine						
C_{max} (μg/mL)	162 ± 11.0	168 ± 3.88	489 ± 27.7	432	1720 ± 122	1480 ± 226
AUC_{inf} (μg·day/mL)	426 ± 28.1	436 ± 18.4	1520[*2]	1330	–	–
$AUC_{0\text{-}3}$ (μg·day/mL)	206 ± 3.34	198 ± 3.09	683 ± 14.0	598	1510 ± 57.7	1290 ± 110
CL (mL/day/kg)	13.8 ± 0.896	13.5 ± 0.581	13.1[*2]	15.0	–	–
V_c (mL/kg)	36.2 ± 2.44	34.9 ± 0.802	41.6[*2]	45.9	–	–
V_{ss} (mL/kg)	75.8 ± 3.42	79.3 ± 0.629	87.9[*2]	98.8	–	–
MRT (day)	5.49 ± 0.181	5.89 ± 0.286	6.73[*2]	6.59	–	–
$t_{1/2}$ (day)	4.25 ± 0.188	4.44 ± 0.214	5.76[*2]	5.49	–	–
PK parameters of total trastuzumab						
C_{max} (μg/mL)	147 ± 12.1	152 ± 6.21	546 ± 17.6	472	1720 ± 81.9	1510 ± 223
AUC_{inf} (μg·day/mL)	734 ± 78.3	813 ± 87.4	2810[*2]	2500	–	–
$AUC_{0\text{-}3}$ (μg·day/mL)	198 ± 4.52	191 ± 3.86	687 ± 30.5	615	1820 ± 86.8	1500 ± 98.9
CL (mL/day/kg)	8.06 ± 0.817	7.28 ± 0.826	7.04[*2]	7.91	–	–
V_c (mL/kg)	40.2 ± 3.45	38.8 ± 1.61	36.8[*2]	42.0	–	–
V_{ss} (mL/kg)	97.9 ± 6.00	102 ± 4.40	106[*2]	115	–	–
MRT (day)	12.3 ± 1.79	14.1 ± 2.00	15.1[*2]	14.5	–	–
$t_{1/2}$ (day)	9.08 ± 1.28	10.3 ± 1.43	11.3[*2]	10.8	–	–
PK parameters of free DM1						
C_{max} (ng/mL)	13.2 ± 2.11	8.27 ± 0.858	43.9 ± 1.38	59.6	124 ± 12.9	154 ± 13.9
AUC_{all} (ng·day/mL)	3.87 ± 0.387	1.62 ± 0.242	17.3 ± 3.23	36.5	51.6 ± 4.18	78.2 ± 7.53

Arithmetic mean ± SD, n = 3, *1: PK parameters were calculated by a 2-compartment model for trastuzumab emtansine and total trastuzumab, and PK parameters for free DM1, by a model-independent analysis for DM1.
*2: Only the mean value is given because n = 2.

DM1 (0.05, 0.1, 0.2 mg/kg) was administered intravenously in a single dose to male and female rats, and plasma concentration of free DM1 was measured (the table below). No clear sex differences were observed in PK parameters of free DM1. CL of free DM1 decreased with an increase in dose, with AUC_{inf} showing a tendency to increase more than dose-proportionally. In contrast, C_{max} and $AUC_{0\text{-}180}$ increased dose-proportionally.

The applicant explained these results as follows:
Since the last blood sampling point with quantifiable level of plasma free DM1 varied among doses, the calculated apparent elimination half-life tended to be lower than expected in the low dose groups, resulting in an apparent tendency of more than dose-proportional increase in AUC_{inf}. However, C_{max} and $AUC_{0\text{-}180}$ increased dose-proportionally. When these results are taken into account, the PK of DM1 in rats is considered to show a linear response within the dose range from 0.05 to 0.2 mg/kg. Also, V_c and V_{ss} of DM1 were higher compared with the body water content in rats (668 mL/kg) (*Pharm Res.* 1993;10:1093-5), which suggested a high tissue distribution of DM1.

PK parameters of free DM1 after single intravenous administration of DM1 to male and female rats*

Does of DM1 (mg/kg)	0.05		0.1		0.2	
Sex	Male	Female	Male	Female	Male	Female
C_{max} (ng/mL)	10.0 ± 1.99	8.49 ± 0.933	16.6 ± 2.70	16.3 ± 2.19	37.5 ± 2.11	38.5 ± 8.37
AUC_{inf} (ng·min/mL)	1100 ± 447	728 ± 347	1660 ± 99.5	2300 ± 702	6180 ± 1020	7000 ± 435
$AUC_{0\text{-}180}$ (ng·min/mL)	343 ± 34.1	308 ± 27.0	576 ± 42.0	637 ± 66.3	1320 ± 49.2	1540 ± 179
CL (mL/day/kg)	53.5 ± 25.2	79.4 ± 29.0	57.3 ± 2.54	44.0 ± 10.8	32.9 ± 5.69	28.5 ± 2.34
V_c (mL/kg)	5170 ± 939	6000 ± 862	5820 ± 776	5930 ± 837	5320 ± 332	5370 ± 1330
V_{ss} (mL/kg)	21,500 ± 1280	22,500 ± 1680	25,200 ± 2120	25,000 ± 1680	27,100 ± 741	23,500 ± 3860
$t_{1/2}$ (min)	335 ± 138	242 ± 127	317 ± 28.8	429 ± 131	599 ± 93.3	590 ± 82.2

Arithmetic mean ± SD, n = 4, *: PK parameters were calculated by a 2-compartment model.

MCC-DM1 (0.1, 0.5 mg/kg) was administered intravenously in a single dose to male and female rats, and plasma concentrations of DM1 and MCC-DM1 were measured. CL of MCC-DM1 was higher than hepatic blood flow rate in rats (55.2 mL/min/kg) (*Handbook of Essential Pharmacokinetics, Pharmacodynamics and Drug Metabolism for Industrial Scientists.* [Bioneer Life Science 2001, CA, USA]). The applicant explained that the results suggested the contribution of extrahepatic clearance to the elimination of MCC-DM1. V_c of MCC-DM1 was similar to the body water content. The conversion rate from MCC-DM1 to DM1 was low, being approximately 1% in both groups (0.844% and 1.12% in the 0.1 and 0.5 mg/kg groups, respectively). The applicant explained that these data suggested that, after intravenous administration, MCC-DM1 was excreted from the body as unchanged MCC-DM1 or metabolites other than DM1.

Lys-MCC-DM1 (0.3, 0.9 mg/kg) was administered intravenously in a single dose to female rats, and plasma concentrations of DM1, MCC-DM1, and Lys-MCC-DM1 were measured. In the 0.9 mg/kg group, CL and $t_{1/2}$ of Lys-MCC-DM1 were 71.9 mL/min/kg and 7.3 minutes, respectively, with CL of Lys-MCC-DM1 being higher than the hepatic blood flow rate in rats. The applicant explained that these results suggested the contribution of extrahepatic clearance to the elimination of Lys-MCC-DM1. V_c of Lys-MCC-DM1 was 399 mL/kg, which was similar to the body water content in rats. From the observations that little or no MCC-DM1 was detected in the plasma, and that the conversion rate from Lys-MCC-DM1 to DM1 was as low as 0.993%, the applicant explained that Lys-MCC-DM1 was excreted from the body as unchanged Lys-MCC-DM1 or metabolites other than DM1 and MCC-DM1.

Trastuzumab emtansine (3, 10, 30 mg/kg) was administered intravenously in a single dose to male and female cynomolgus monkeys, and serum concentrations of trastuzumab emtansine and total trastuzumab and plasma concentration of free DM1 were measured (the table below). After the administration, trastuzumab emtansine was eliminated in a biphasic manner. CL of trastuzumab emtansine and total trastuzumab was lower in the 10 mg/kg group than in the 3 mg/kg group but comparable between the 10 mg/kg group and the 30 mg/kg group. CL of trastuzumab emtansine was 1.5 to 2 times that of total trastuzumab. V_c of trastuzumab emtansine and total trastuzumab was similar to the plasma volume in monkeys (44.7 mL/kg) (*Handbook of Essential Pharmacokinetics, Pharmacodynamics and Drug Metabolism for Industrial Scientists.* [Bioneer Life Science 2001, CA, USA]). No clear sex differences were observed in the PK of trastuzumab emtansine or total trastuzumab.

Plasma free DM1 concentration peaked immediately after the administration, then decreased with time. C_{max} and AUC_{all} of free DM1 increased with the dose of trastuzumab emtansine. The exposure level of free DM1 tended to be slightly higher in males than in females. From the findings that C_{max} and AUC of free DM1 after the first dose in repeated intravenous administration in cynomolgus monkeys tended to be slightly higher in females than in males, and that a consistent

tendency was not observed between the single dose study and the repeat dose study, the applicant explained that the higher levels of free DM1 observed in males in the single dose study was likely to be due to inter-individual variability and not due to sex difference.

PK parameters after single intravenous administration of trastuzumab emtansine to male and female cynomolgus monkeys*[1]

Dose of trastuzumab emtansine (mg/kg)	3		10		30	
Sex	Male	Female	Male	Female	Male	Female
PK parameters of trastuzumab emtansine						
C_0 (μg/mL)	84.4 ± 11.2	75.1 ± 4.92	278 ± 16.8	257 ± 36.2	784 ± 66.3	680 ± 66.8
AUC_{inf} (μg·day/mL)	194 ± 19.0	203 ± 19.2	918 ± 68.6	844 ± 121	2770 ± 296	3090 ± 693
CL (mL/day/kg)	15.7 ± 1.59	15.0 ± 1.36	10.8 ± 0.772	11.8 ± 1.59	10.7 ± 1.21	9.86 ± 2.38
V_c (mL/kg)	36.4 ± 5.05	40.5 ± 2.62	35.5 ± 2.07	38.7 ± 5.10	37.7 ± 3.33	43.5 ± 4.44
$t_{1/2}$ (day)	2.53 ± 0.106	2.68 ± 0.205	4.40 ± 0.491	3.86 ± 0.452	4.90 ± 1.09	5.44 ± 0.200
PK parameters of total trastuzumab						
C_0 (μg/mL)	88.0 ± 9.55	81.9 ± 3.42	280 ± 13.7	253 ± 23.1	743 ± 71.2	697 ± 59.9
AUC_{inf} (μg·day/mL)	301 ± 25.0	325 ± 33.4	1630 ± 145	1430 ± 231	5340 ± 1070	5580 ± 1140
CL (mL/day/kg)	10.1 ± 0.841	9.39 ± 0.915	6.06 ± 0.524	6.98 ± 1.05	5.67 ± 1.29	5.43 ± 1.24
V_c (mL/kg)	34.7 ± 4.00	37.1 ± 1.51	35.2 ± 1.74	39.1 ± 3.53	39.8 ± 3.98	42.3 ± 3.81
$t_{1/2}$ (day)	4.39 ± 0.328	4.58 ± 0.410	8.53 ± 1.08	7.40 ± 0.960	10.9 ± 3.29	9.91 ± 0.240
PK parameters of free DM1						
C_{max} (ng/mL)	7.32 ± 0.252[*2]	6.87 ± 0.877[*2]	26.4 ± 2.60	20.4 ± 2.07	69.0 ± 3.21	50.7 ± 8.99
AUC_{all} (ng·day/mL)	–	–	29.9 ± 2.24	20.4 ± 7.41	83.9 ± 12.1	72.2 ± 7.59

Arithmetic mean ± SD, n = 3, *1: PK parameters were calculated by a model-independent analysis. *2: Plasma concentration at the first blood sampling point

3.(ii).A.(2).2) Repeat-dose administration

Trastuzumab emtansine (1, 3, 10 mg/kg) was administered intravenously once every 3 weeks for a total of 8 doses to male and female cynomolgus monkeys, and serum concentrations of trastuzumab emtansine and total trastuzumab and plasma concentration of free DM1 were measured (the table below).

After both the first and the eighth doses, C_{max} of trastuzumab emtansine and total trastuzumab increased in a generally dose-proportional manner, whereas CL decreased with increasing doses, and AUC tended to increase more than dose-proportionally. The applicant explained that high-dose trastuzumab emtansine may have saturated the antigen-dependent elimination pathway mediated by the binding of trastuzumab emtansine to HER2, which resulted in the non-linear PK of trastuzumab emtansine and total trastuzumab.

C_{max} of free DM1 increased in a generally dose-proportional manner, whereas AUC increased more than dose-proportionally. The applicant explained that these results were likely due to the limited number of blood sampling points with a quantifiable level of free DM1 in the low dose group. AUC of trastuzumab emtansine after the eighth doses was 1.2, 1.4, and 1.5 times that observed after the first dose in the 1, 3, and 10 mg/kg groups, respectively, and AUC of total trastuzumab was 1.3, 1.7, and 2.1 times, respectively, showing a tendency of the accumulation of trastuzumab emtansine and total trastuzumab after repeated administration of trastuzumab emtansine in the 10 mg/kg group. AUC of free DM1 after the eighth dose in the 10 mg/kg group was 1.6 times that observed after the first dose, also showing a tendency of DM1 accumulation after repeated administration of trastuzumab emtansine in the 10 mg/kg group. The applicant explained that, due to the limited number of time points with quantifiable level of free DM1, it was practically impossible to evaluate the effect of repeated administration on the PK of free DM1 in the 1 and 3 mg/kg groups.

PK parameters after repeated intravenous administration of trastuzumab emtansine to male and female monkeys (once every 3 weeks for a total of 8 doses) *[1]

Dose of trastuzumab emtansine (mg/kg)	1		3		10	
Administration	Cycle 1	Cycle 8	Cycle 1	Cycle 8	Cycle 1	Cycle 8
PK parameters of trastuzumab emtansine						
C_{max} (μg/mL)	21.9 ± 2.03	28.3 ± 5.85	71.7 ± 5.40	90.3 ± 9.79	190 ± 37.0	257 ± 41.9
AUC (μg·day/mL)*[2]	33.4 ± 3.44	39.8 ± 8.62	149 ± 20.3	212 ± 36.7	646 ± 83.6	973 ± 145
CL (mL/day/kg)	30.3 ± 3.38	26.4 ± 6.84	20.4 ± 2.77	14.5 ± 2.79	15.0 ± 2.12	9.94 ± 1.61
$t_{1/2}$ (day)	1.93 ± 0.139	1.82 ± 0.561	2.54 ± 0.251	3.03 ± 0.441	4.71 ± 0.527	4.55 ± 1.23
PK parameters of total trastuzumab						
C_{max} (μg/mL)	21.3 ± 1.68	29.4 ± 3.95	68.2 ± 4.89	84.4 ± 9.16	187 ± 32.2	330 ± 48.2
AUC (μg·day/mL)*[2]	41.3 ± 4.77	52.1 ± 13.5	193 ± 28.5	319 ± 75.9	903 ± 111	1870 ± 370
CL (mL/day/kg)	24.4 ± 3.08	20.4 ± 6.40	15.6 ± 2.44	9.40 ± 2.63	9.62 ± 1.49	4.45 ± 1.50
$t_{1/2}$ (day)	2.89 ± 0.365	2.88 ± 1.19	3.83 ± 0.497	5.06 ± 1.36	7.61 ± 1.17	9.42 ± 2.70
PK parameters of free DM1						
C_{max} (ng/mL)	1.56 ± 0.221	1.86 ± 1.22	3.64 ± 0.452	3.45 ± 0.534	13.0 ± 3.88	11.2 ± 1.94
AUC (ng·day/mL)	0.886*[3]	0.399 ± 0.0426	1.21 ± 0.254	1.92 ± 1.91	7.57 ± 1.78	12.1 ± 4.73
$t_{1/2}$ (day)	0.524*[3]	0.201 ± 0.0172	0.518 ± 0.208	1.03 ± 1.27	2.05 ± 0.447	3.52 ± 1.28

Arithmetic mean ± SD, n = 12 (6/sex), *1: PK parameters were calculated by a model-independent analysis.
*2: $AUC_{0\text{-}21}$ in Cycle 1, $AUC_{147\text{-}168}$ in Cycle 8,*3: n = 1

Trastuzumab emtansine (3, 10, 30 mg/kg) was administered intravenously once every 3 weeks for a total of 4 doses. As a result, CL of trastuzumab emtansine and total trastuzumab after the fourth dose was lower in the 10 mg/kg group than in the 3 mg/kg group but similar between the 10 mg/kg group and the 30 mg/kg group. C_{max} of free DM1 increased in a generally dose-proportional manner within the dose range from 3 to 30 mg/kg. AUC_{all} increased more than dose-proportionally between the 3 mg/kg and the 10 mg/kg groups, and in a generally dose-proportional manner between the 10 mg/kg and the 30 mg/kg groups.

On the basis of the above results, the applicant explained that, when trastuzumab emtansine (1-30 mg/kg) was administered repeatedly to monkeys, the PK of trastuzumab emtansine, total trastuzumab, and free DM1 is likely to be linear at ≥10 mg/kg.

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

3.(ii).A.(3).1) Tissue distribution

i) Trastuzumab emtansine or trastuzumab

Trastuzumab emtansine containing ^{125}I-labeled trastuzumab ([^{125}I]trastuzumab-MCC-DM1) or ^{125}I-labeled trastuzumab was administered at 13 mg/kg intravenously in a single dose to female rats, and tissue distribution of radioactivity was investigated by quantitative whole body autoradiography (QWBA). The tissue distribution of radioactivity was similar between animals receiving ^{125}I-labeled trastuzumab and animals receiving [^{125}I]trastuzumab-MCC-DM1, from which the applicant explained that the distribution of trastuzumab emtansine in the body reflects the behavior characteristic to trastuzumab molecules. The highest radioactivity was observed in blood at all time points, and radioactivity was also observed in tissues with abundant blood flow (e.g., lung, liver, kidney, heart, spleen). Radioactivity in tissues decreased over time with the decrease in the radioactivity in blood, showing no tendency to remain in tissues.

Trastuzumab emtansine containing ^{3}H-labeled DM1 (trastuzumab-MCC-[^{3}H]DM1) was administered at 13 mg/kg intravenously in a single dose to female rats, and tissue distribution of radioactivity was investigated by QWBA. The highest radioactivity was observed in plasma at all time points. The percentage of injected radioactivity dose per gram tissue (%ID/g tissue) at 1 hour

after administration was the highest in the plasma, followed in descending order by the liver, kidney, lung, spleen, heart, bone marrow, and thymus (12%ID/g tissue, 2.8%ID/g tissue, 2.0%ID/g tissue, 1.6%ID/g tissue, 1.6%ID/g tissue, 0.93%ID/g tissue, 0.84%ID/g tissue, and 0.18%ID/g tissue, respectively). Radioactivity in the kidney and thymus hardly decreased within 7 days after administration, but radioactivity in all tissues decreased by 14 days after administration, showing no tendency of remaining in tissues. Tissue distribution of radioactivity was similar between trastuzumab-MCC-[^{3}H]DM1 and [^{125}I]trastuzumab-MCC-DM1.

ii) DM1

Trastuzumab-MCC-[^{3}H]DM1 (0.2 mg/kg) was administered intravenously in a single dose to female rats, and tissue distribution of radioactivity was investigated by QWBA. Radioactivity in blood decreased rapidly until the following day of administration, after which it disappeared slowly. At all time points, radioactivity in most tissues was higher than that in the blood. Radioactivity at 10 minutes after administration in the lung, kidney, liver, spleen, adrenal, and heart was 3.07%ID/g tissue, 2.88%ID/g tissue, 1.89%ID/g tissue, 1.77%ID/g tissue, 1.77%ID/g tissue, and 1.13%ID/g tissue, respectively. The applicant explained that these results suggested the high tissue distribution of DM1. Radioactivity in tissues was 10 to 30 times higher than that in the blood, but did not show any tendency to remain.

3.(ii).A.(3).2 Plasma protein binding and distribution in blood cells

DM1 (20, 100, 1000 ng/mL) was incubated with rat, monkey, and human plasma samples, and plasma protein binding rate was investigated by the equilibrium dialysis method. Plasma protein binding rate of DM1 was 96.6% to 97.5% in rats, 90.5% to 92.2% in monkeys, and 91.8% to 93.2% in humans, showing an almost constant rate within the concentration range studied in all animal species tested.

Although distribution of DM1 in human blood cells was not investigated, the applicant explained that DM1 was likely to be distributed in blood cells, based on the findings that radioactivity distribution was similar between plasma fraction and blood cell fraction in the tissue distribution study in rats using ^{3}H-labeled DM1.

3.(ii).A.(3).3 Placental and fetal transfer

Placental and fetal transfer of trastuzumab emtansine was not investigated. However, the applicant explained that administration of trastuzumab emtansine should be avoided in pregnant women or in women who may possibly be pregnant, because trastuzumab, the main component of trastuzumab emtansine, was shown to be transferred to amniotic fluid and fetuses (see package inserts of Herceptin Intravenous Infusion 60 and Herceptin Intravenous Infusion 150).

3.(ii).A.(4) Metabolism

3.(ii).A.(4).1 Trastuzumab emtansine

i) Stability in plasma

Trastuzumab-MCC-[^{3}H]DM1 or unlabeled trastuzumab emtansine (50 μg/mL each) was incubated with rat, monkey, and human plasma samples at 37°C for 96 hours to investigate the stability of trastuzumab emtansine in plasma.

In the study using trastuzumab-MCC-[^{3}H]DM1, following deproteinization of the incubated samples, approximately 80% of the added radioactivity was found in the precipitate in all animal species studied. The applicant explained that these results suggested that DM1 was present mostly in protein-bound form in plasma.

In the study using unlabeled trastuzumab emtansine, free DM1 concentration in plasma was 3.79 to 5.30 ng/mL immediately after the initiation of incubation, then increased to 105 to 118 ng/mL after 96 hours of incubation. The applicant explained that the results suggested that DM1 was

released from trastuzumab emtansine over time.

ii) *In vivo* metabolism

Trastuzumab-MCC-[^{3}H]DM1 (10, 13 mg/kg) was administered intravenously in a single dose to female rats, and metabolites of trastuzumab emtansine in plasma were investigated. Following deproteinization of the obtained samples, ≥95% of plasma radioactivity was found in the precipitate. The applicant explained that these results, along with the results of the SE-HPLC and ELISA, suggested that trastuzumab emtansine was present mainly as the unchanged compound. Free DM1 (corresponding to 0.3% of $AUC_{0\text{-}168}$ of radioactivity in plasma) was detected only when a reducing agent was added to the supernatant obtained after deproteinization. The applicant explained that these results suggested that DM1 was present in a disulfide-bound form in plasma.

Trastuzumab emtansine (10 mg/kg) was administered intravenously in a single dose to female rats, and plasma concentrations of free DM1, MCC-DM1, and Lys-MCC-DM1 were measured. The mean plasma concentration (range) of free DM1 was 10.49 ng/mL (6.36-15.56 ng/mL) at 15 minutes after administration, which decreased to 1.89 ng/mL (1.56-2.20 ng/mL) at 7 days after administration. Plasma MCC-DM1 concentrations were below the lower limit of quantitation (3.81 ng/mL) at all time points up to 7 days after administration, except in 1 animal (9.94 ng/mL) at 7 days after administration for which it was quantifiable. Plasma Lys-MCC-DM1 concentrations were quantifiable (5.44-15.43 ng/mL) in 5 of 6 animals at 1 day after administration, but fell below the lower limit of quantitation (2.15 ng/mL) in 5 of 6 animals at 5 days after administration and in 3 of 6 animals at 7 days after administration.

Trastuzumab-MCC-[^{3}H]DM1 (10 mg/kg) was administered intravenously in a single dose to untreated female rats and to biliary cannulated female rats, and metabolites of trastuzumab emtansine in urine, feces, and bile were investigated. When samples were deproteinized, 94.6% to 99.5% and 78.4% to 86.3% of radioactivity recovered in urine and feces, respectively, in untreated rats, and 96.9% to 99.5% of radioactivity recovered in the bile of biliary cannulated rats, were found in the supernatant liquid of the samples. The applicant explained that these results suggested that little or no unchanged trastuzumab emtansine was present in urine, feces, or bile. When no reducing agent was added, MCC-DM1 and Lys-MCC-DM1 were detected as the main metabolites in urine and bile up to 6 days after the administration (the amounts of the two metabolites detected in urine were 0.2% and 2.2%, respectively, of the administered radioactivity; and in bile, 4.6% and 21.5%, respectively, of the administered radioactivity). Only when a reducing agent was added, a trace amount of free DM1 was detected in urine and bile (0.1% and 1%, respectively, of the administered radioactivity). The applicant explained that these results suggested that DM1 in urine and bile was present mostly in a disulfide-bound form. $[^3H]_2O$ was detected as a metabolite in urine and bile (2.3% and 3.6%, respectively, of the administered radioactivity); there were no other metabolites with the content exceeding 1% of the administered radioactivity.

3.(ii).A.(4).2) DM1

i) *In vitro* metabolism

DM1 (1 μmol/L) was incubated with human liver microsomes or with recombinant human CYP (1A2, 2A6, 2B6, 2C8, 2C9, 2C18, 2C19, 2D6, 2E1, 3A4, 3A5, 4A11) at 37°C for 60 minutes, and formation of DM1 metabolites was investigated.

In the study using human liver microsomes, the residual rate of DM1 in the presence and absence of reduced nicotinamide-adenine dinucleotide phosphate (NADPH) was 26.8% and 82.6%, respectively. When 1-aminobenzotriazole (a non-selective CYP inhibitor) was added, the residual rate of DM1 in the presence of NADPH increased to 81.2%. The applicant explained that these results suggested that NADPH was required for the metabolism of DM1 and that CYP is mainly involved in the metabolism.

In the study using human liver microsomes, addition of ketoconazole and troleandomycin (inhibitors of CYP3A4/5) increased the residual rate of DM1 to 76.5% and 84.2%, respectively, in the presence of NADPH, whereas inhibitors of other CYP isoforms (CYP1A2, 2A6, 2B6, 2C19, 2C8, 2C9, 2D6) had no clear effect on the metabolism of DM1 (residual rates, 22.0%-27.9%). In the study using recombinant CYP, DM1 was metabolized by CYP3A4-expressing system (residual rate, 21.7%) and CYP3A5-expressing system (residual rate, 54.5%), but hardly metabolized by other CYP-expressing systems (residual rates, 76.3%-112%).

The applicant explained that these results suggested that DM1 was metabolized mainly by CYP3A4 and partly by CYP3A5.

ii) *In vivo* metabolism

^{3}H-labeled DM1 (0.2 mg/kg) was administered intravenously in a single dose to biliary cannulated female mice, and metabolites of trastuzumab emtansine in bile and plasma were investigated. In the bile collected up to 24 hours after the administration, M3 to M6 (DM1 metabolites with oxidized or methylated thiol groups) were detected (3.3%-7.2% of the administered radioactivity). When no reducing agent was added to the bile samples, M9, M10, and M11, which are derivatives of glutathione conjugates, were slightly detected. When a reducing agent was added, none of these metabolites were detected; instead, DM1 was detected in an amount corresponding to 1.9% of the administered radioactivity. The applicant explained that DM1 detected in the presence of the reducing agent was the sum of DM1 (monomer and dimer) present in the sample and DM1 generated by reduction of M9, M10, and M11.

A trace amount of M3 was detected as the only metabolite in plasma.

3.(ii).A.(5) Excretion

3.(ii).A.(5).1) Urinary, fecal, and biliary excretion

Trastuzumab-MCC-[^{3}H]DM1 (10, 13 mg/kg) was administered intravenously in a single dose to female rats, and urinary and fecal excretion rates of radioactivity (% of dose) were investigated. Following administration of trastuzumab-MCC-[^{3}H]DM1 at 10 mg/kg, the urinary and fecal excretion rates of radioactivity up to 7 days after administration were 8.2% and 50%, respectively. Following administration of trastuzumab-MCC-[^{3}H]DM1 at 13 mg/kg, the urinary and fecal excretion rates of radioactivity up to 14 days after administration were 2.3% and 63%, respectively. When urine and fecal samples were deproteinized, 94.6% to 99.5% and 78.4% to 86.3% of radioactivity recovered in urine and feces, respectively, were found in the supernatant.

Trastuzumab-MCC-[^{3}H]DM1 (10 mg/kg) was administered intravenously in a single dose to biliary cannulated female rats, and biliary and fecal excretion rates of radioactivity (% of the administered radioactivity) were investigated. Biliary and fecal excretion rates of radioactivity up to 7 days after administration were 51% and 4.7%, respectively. When biliary samples were deproteinized, 96.9% to 99.5% of radioactivity recovered in the bile was found in the supernatant.

The applicant explained that these results suggested that, follwoing intravenous administration of trastuzumab emtansine to rats, DM1 moiety of trastuzumab emtansine was mostly excreted into feces via bile in the form of free DM1 or its metabolites. It was also suggested that, following single intravenous administration of ^{3}H-labeled DM1 to rats, DM1 was excreted in feces mainly via the bile.

3.(ii).A.(5).2) Excretion in milk

Excretion of trastuzumab emtansine in milk was not investigated. However, the applicant explained that breast feeding should be avoided during treatment with trastuzumab emtansine because trastuzumab, the main component of trastuzumab emtansine, is shown to be excreted in

milk (see package inserts of Herceptin Intravenous Infusion 60 and Herceptin Intravenous Infusion 150).

3.(ii).A.(6) Pharmacokinetic interactions

In the regulatory submission of trastuzumab emtansine, non-clinical data on pharmacokinetic interaction studies using DM1 were submitted, but data on studies using trastuzumab emtansine were not.

The applicant explained the reasons for conducting nonclinical pharmacokinetic interaction studies on DM1 but not on trastuzumab emtansine, as follows:
(i) CYPs are not considered to be involved in the catabolic metabolism of trastuzumab emtansine, and (ii) DM1 was detected in plasma following intravenous administration of trastuzumab emtansine to various animal species and to humans.

3.(ii).A.(6).1) Enzyme inhibition

Substrates of CYP isoforms (CYP1A2, 2B6, 2C8, 2C9, 2C19, 2D6, 3A4) were incubated with human liver microsomes in the presence of DM1 (1.4-678 nmol/L). As a result, DM1 did not show any inhibitory effect on CYP isoforms even at 678 nmol/L, the maximum concentration studied.

A substrate of CYP3A4 was preincubated with human liver microsomes for 30 minutes in the presence of DM1 (1.4-678 μmol/L). DM1 inhibited CYP3A4 in a time-dependent manner, with IC_{50} of 155 nmol/L (114 ng/mL).

These results showed that DM1 inhibited CYP3A4 in a time-dependent manner. However, the applicant explained that, following administration of trastuzumab emtansine, free DM1 is unlikely to cause pharmacokinetic interactions with CYP3A4 through the time-dependent inhibition, for the following reasons.

- In the Japanese and foreign clinical studies in which trastuzumab emtansine was administered to patients with breast cancer according to the proposed dosage regimen (3.6 mg/kg intravenous infusion, every 3 weeks), the mean C_{max} of free DM1 in Cycle 1 of treatment was 3.41 to 5.42 ng/mL.

- In global phase III studies (Study TDM4370g/Study BO21977, [EMILIA study]) (287 subjects included in PK analysis), the maximum C_{max} of free DM1 throughout all treatment cycles was 59.7 ng/mL.

- When trastuzumab emtansine was administered according to the proposed dosage regimen, there was no tendency of accumulation of free DM1 after Cycle 1.

3.(ii).A.(6).2) Enzyme induction

Cryopreserved human hepatocytes were treated with DM1 (0.1-1000 nmol/L) for 48 hours, and the mRNA expression levels and enzyme activities of CYP1A2, 2B6, and 3A4/5 were investigated. DM1 did not affect cell viability within the concentration range investigated, nor did it increase any mRNA expression level or enzyme activity of CYP1A2, 2B6, or 3A4/5. On the basis of these results, the applicant explained that DM1 is unlikely to cause pharmacokinetic interactions through induction of CYP1A2, 2B6, or 3A4/5.

3.(ii).A.(6).3) Transporters

P-gp-mediated transport of ^{3}H-labeled DM1 (0.5 μmol/L) was investigated using MDCK II cell lines derived from dog kidney cells engineered to express human P-gp (MDCK II-MDR1 cell

lines). The apparent efflux ratio (apparent permeability coefficient from basolateral surface to apical surface [$P_{app\ B\rightarrow A}$]/apparent permeability coefficient from apical surface to basolateral surface [$P_{app\ A\rightarrow B}$]) was 0.89 and 6.65 in MDCK II cell lines and MDCK II-MDR1 cell lines, respectively. The DM1 efflux ratio ($P_{app\ B\rightarrow A}/P_{app\ A\rightarrow B}$) in MDCK II-MDR1 cell lines, adjusted with the data of MDCK II cell lines, was 7.47. The apparent efflux ratio ($P_{app\ B\rightarrow A}/P_{app\ A\rightarrow B}$) of DM1 in MDCK II-MDR1 cell lines was 1.10 and 1.76 in the presence of P-gp inhibitor PSC833 and verapamil, respectively. The values were lower than that observed in their absence (5.90).

In a preliminary study using a human breast cancer-derived cell lines engineered to express human breast cancer-resistant protein (SK-BR-3/BCRP), DM1 was shown not to serve as a substrate for BCRP.

Inhibitory effect of trastuzumab emtansine (0.5 μmol/L) on P-gp-mediated digoxin transport was investigated using MDCK II-MDR1 cell lines. The apparent efflux ratio ($P_{app\ B\rightarrow A}/P_{app\ A\rightarrow B}$) of digoxin was 9.90 and 9.65 in the absence and presence of DM1, respectively, showing that DM1 did not inhibit P-gp.

3.(ii).A.(7) ATA production in monkeys

In single and repeat dose studies of trastuzumab emtansine in male and female monkeys, ATA was detected in the serum in 8 of 190 animals (4.2%).

In the repeated intravenous dose study (once every 3 weeks for a total of 8 doses), ATA-positive cases were observed most frequently (3 animals in the 1 mg/kg group, 1 animal in the 10 mg/kg group). Therefore, the effect of ATA on the PK of trastuzumab emtansine was investigated based on the results of the study. In the 1 mg/kg group, PK parameters of trastuzumab emtansine, total trastuzumab, and free DM1 in ATA-positive animals, ATA-negative animals, and all animals were as shown in the table below. No clear differences were observed in C_{max} of trastuzumab emtansine, total trastuzumab, or free DM1 between ATA-positive group and ATA-negative group, whereas AUC of trastuzumab emtansine and total trastuzumab in ATA-positive group was approximately 60% of that of the ATA-negative group.

The applicant explained the above results as follows:
The results suggested that ATA may affect the PK of trastuzumab emtansine although no definite conclusion can be derived at present because of the limited number of animals studied. However, based on the low incidence of ATA-positive animals and the observation that ATA-positive animals were observed mostly in the low dose group in the toxicity study, ATA-positive animals are unlikely to have any significant effect on the interpretation of the results of the toxicity study.

PK parameters after repeated intravenous administration of trastuzumab emtansine to male and female monkeys once every 3 weeks for 8 doses (after 8th dosing)

	ATA-positive animals	ATA-negative animals	All animals
n	3	9	12
PK parameters of trastuzumab emtansine			
C_{max} (μg/mL)	25.5 ± 6.97	29.3 ± 5.55	28.3 ± 5.85
$AUC_{147\text{-}168d}$ (μg·day/mL)	27.7 ± 3.18	43.8 ± 5.18	39.8 ± 8.63
CL (mL/day/kg)	36.4 ± 4.36	23.1 ± 3.06	26.4 ± 6.84
$t_{1/2}$ (day)	1.18 ± 0.243	2.04 ± 0.459	1.82 ± 0.561
PK parameters of total trastuzumab			
C_{max} (μg/mL)	25.7 ± 3.76	30.6 ± 3.34	29.4 ± 3.95
$AUC_{147\text{-}168d}$ (μg·day/mL)	33.8 ± 3.35	58.2 ± 8.89	52.1 ± 13.5
CL (mL/day/kg)	29.8 ± 3.18	17.3 ± 3.07	20.4 ± 6.40
$t_{1/2}$ (day)	1.66 ± 0.175	3.29 ± 1.09	2.88 ± 1.19
PK parameters of free DM1			
C_{max} (ng/mL)	1.35 ± 0.305	2.03 ± 1.38	1.86 ± 1.22

Arithmetic mean ± SD

3.(ii).A.(8) Effect of changes in drug substance manufacturing process on PK

[redacted].

(a)
(b)
(c)
(d)

[redacted]

In order to investigate the effect of changes (a) and (c) above on the PK of trastuzumab emtansine, the formulations prepared from the drug substance before and after the change in the manufacturing process were administered intravenously at 10 mg/kg in a single dose to female rats and female cynomolgus monkeys, and serum trastuzumab emtansine concentrations were measured.

3.(ii).A.(8).1) Comparison between formulations manufactured by processes A and B

PK parameters of trastuzumab emtansine in female monkeys were as shown in the table below. Geometric mean ratios [90% confidence interval (CI)] of AUC_{last} and C_{max} of the formulation manufactured by process B to those of the formulation manufactured by process A were 0.830 [0.732, 0.940] and 0.952 [0.880, 1.03], respectively, showing that C_{max} was not significantly different between the two formulations, whereas AUC_{last} of the formulation manufactured by process B was approximately 17% lower than that of the formulation manufactured by process A.

PK parameters of trastuzumab emtansine in female monkeys

Drug substance manufacturing process	C_{max} (μg/mL)	AUC_{last} (μg·day/mL)	CL (mL/day/kg)	$t_{1/2}$ (day)	V_{ss} (mL/kg)
Process A	253 ± 28.5	701 ± 134	14.9 ± 3.85	3.79 ± 0.878	73.1 ± 6.10
Process B	241 ± 28.3	575 ± 74.4	17.7 ± 2.72	4.28 ± 0.752	87.8 ± 8.21

Arithmetic mean ± SD, n = 13

In rats, the geometric mean ratios [90% CI] of AUC_{last} and C_{max} of the formulation manufactured by process B to those of the formulation manufactured by process A were 0.923 [0.878, 0.971] and 0.911 [0.869, 0.956], respectively, showing no clear differences in the PK parameter of trastuzumab emtansine between the two formulations.

3.(ii).A.(8).2) Comparison between formulations manufactured by processes C and D

PK parameters of trastuzumab emtansine in female monkeys were as shown in the table below. Geometric mean ratios [90% CI] of AUC_{last} and C_{max} of the formulation manufactured by process D to those of the formulation manufactured by process C were 0.983 [0.889, 1.09] and 0.820 [0.761, 0.883], respectively, showing that AUC_{last} was not significantly different between the two formulations, whereas C_{max} of the formulation manufactured by process D was approximately 18% lower than that of the formulation manufactured by process C.

PK parameters of trastuzumab emtansine in female monkeys

Drug substance manufacturing process	C_{max} (μg/mL)	AUC_{last} (μg·day/mL)	CL (mL/day/kg)	$t_{1/2}$ (day)	V_{ss} (mL/kg)
Process C	280 ± 24.9	869 ± 141	11.8 ± 2.07	4.50 ± 0.663	64.9 ± 6.34
Process D	230 ± 29.8	850 ± 112	11.9 ± 1.53	4.16 ± 0.825	64.4 ± 8.62

Arithmetic mean ± SD, n = 13

Thus, no clear differences were observed in the PK of trastuzumab emtansine between the two formulations in the study in rats, whereas, a difference was observed in the exposure level (AUC_{last} or C_{max}) of trastuzumab emtansine between the formulations in studies in monkeys.

The applicant explained the above results as follows:
It is unclear why difference in the exposure level of trastuzumab emtansine was observed between the two formulations only in monkeys. However, the extent of the difference in the exposure level was within the range of inter-individual variability of the exposure level observed in the foreign phase III study (Study TDM4370g) (coefficient of variation of C_{max} and AUC_{inf}: 19.8% and 24.9%, respectively, in Cycle 1; 39.3% and 26.7%, respectively, in Cycle 4), which suggested that the differences were not pharmacokinetically significant. Therefore, the changes in the manufacturing process of the drug substance are unlikely to significantly affect the PK of trastuzumab emtansine.

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

Based on the submitted data and the following review, PMDA concluded that the applicant's explanation on absorption, distribution, metabolism, and excretion of trastuzumab emtansine is acceptable.

Metabolism of trastuzumab emtansine in plasma

DM1 was shown to be released from trastuzumab emtansine over time [see "3.(ii).A.(4).1).(i) Stability in plasma"]. Therefore, PMDA asked the applicant to explain the mechanism of the release of DM1 from trastuzumab emtansine in the plasma.

The applicant responded as follows:
Although the mechanism of the release of DM1 from trastuzumab emtansine is unclear at present, there are the following 3 possible mechanisms.

- The linker is oxidized in the plasma, which destabilizes the thioether bond between DM1 and the linker, resulting in the cleavage of the thioether bond under reducing conditions.
- Exchange of maleimide with active thiol groups in plasma (albumin, free cysteine, glutathione, etc.) (*Bioconjug Chem.* 2011;22:1946-53).
- DM1 bound to a cysteine residue of trastuzumab moiety of trastuzumab emtansine via disulfide bond is released under reducing conditions.

PMDA considers that since no data have been available on the mechanism of the release of DM1 from trastuzumab emtansine and the applicant's discussion remains a matter of speculation, information should be collected continuously.

3.(iii) Summary of toxicology studies

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

The following toxicity studies on trastuzumab emtansine were conducted: studies in cynomolgus monkeys to investigate the toxicity mediated by the binding of trastuzumab emtansine to HER2 (HER2-dependent toxicity) and the toxicity not mediated by the binding of trastuzumab emtansine to HER2 (HER2-independent toxicity), and studies in rats to investigate the toxicity not mediated by the binding of trastuzumab emtansine to HER2 and the toxicity of DM1.

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

3.(iii).A.(1).1) Single intravenous dose study in rats

Trastuzumab emtansine (0 [vehicle], 6, 20, 60 mg/kg) was administered intravenously in a single dose to SD rats (10 each of males and females per group), and 5 each of males and females per group were necropsied 3 days after the administration and all remaining animals 22 days after administration. In the 60 mg/kg group, 2 males and 1 female died or euthanized 5 days after administration, and all remaining animals were euthanized 6 days after the administration because of the aggravation of general conditions.

Tests performed 3 days after administration showed the following findings: (a) hematology findings included increased neutrophil count and decreased platelet count (≥20 mg/kg groups) and decreased reticulocyte count and decreased lymphocyte count (60 mg/kg group); (b) clinical chemistry findings included increased alanine aminotransferase (ALT), increased aspartate aminotransferase (AST), increased alkaline phosphatase (ALP), increased globulin, decreased albumin/globulin ratio (A/G ratio) (≥20 mg/kg groups), increased total bilirubin, increased γ-glutamyl transpeptidase (GGT), increased cholesterol, and decreased glucose (60 mg/kg group); and (c) histopathological findings included increased mitotic figure count in multiple organs, hypertrophy and vacuolization of hepatic Kupffer cells (trastuzumab emtansine groups), degeneration and necrosis of hepatocytes, depletion and necrosis of splenic lymphocytes, necrosis of thymic lymphocytes, degeneration and necrosis of renal tubules, decreased bone marrow cell count, necrosis of mammary gland (≥20 mg/kg groups), degeneration and necrosis of duodenal crypt epithelium, degeneration of testicular seminiferous tubules, cell debris in the ductus epididymis, and necrosis of corpora lutea in the ovary (60 mg/kg group). The test performed 22 days after administration showed that the toxic findings observed in the 6 and 20 mg/kg groups were reversible.

Based on the above results, the highest non-severely toxic dose (HNSTD) was determined to be 20 mg/kg. The exposure levels (AUC_{inf}) at this dose were 1520 μg·day/mL in males and 1330 μg·day/mL in females, which were 4.3 and 3.7 times, respectively, the clinical exposure level.*

*: AUC_{last} in Cycle 1 in Japanese patients receiving trastuzumab emtansine at 3.6 mg/kg every 3 weeks in the Japanese phase II study (Study JO22997)

3.(iii).A.(1).2) Single intravenous dose study in monkeys

Trastuzumab emtansine (0 [vehicle], 3, 10, 30 mg/kg) was administered intravenously in a single dose to cynomolgus monkeys (6/sex/group), and 3 each of males and females per group were necropsied 3 days after administration and all remaining animals 22 days after administration.

Tests performed 3 days after administration showed the following findings: (a) hematology findings included increased neutrophil count, increased monocyte count, increased fibrinogen,

and decreased platelet count (≥10 mg/kg groups), and decreased lymphocyte count (30 mg/kg group); (b) clinical chemistry findings included increased AST and increased ALP (≥10 mg/kg groups), increased globulin, decreased albumin, and decreased A/G ratio (30 mg/kg group); and (c) histopathological findings included hypertrophy of hepatic Kupffer cells and increased mitotic figure count in multiple organs (≥3 mg/kg groups). Tests performed 22 days after administration showed that the toxic findings caused by trastuzumab emtansine recovered or tended to recover, except the toxic findings on albumin and globulin observed in the 30 mg/kg group.

Based on the above results, HNSTD was determined to be 30 mg/kg. The exposure levels (AUC_{inf}) at this dose were 2770 μg·day/mL in males and 3090 μg·day/mL in females, which were 7.8 and 8.7 times, respectively, the clinical exposure level.*

*: AUC_{last} in Cycle 1 in Japanese patients receiving trastuzumab emtansine at 3.6 mg/kg every 3 weeks in the Japanese phase II study (Study JO22997)

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

3.(iii).A.(2).1) Three-week repeat dose study in rats

Trastuzumab emtansine (0 [vehicle], 10, 26, 52 mg/kg) was administered intravenously once weekly for 3 weeks (3 doses in total) to SD rats (5 females per group), and animals were necropsied 28 days after the last dose. Two animals in the 52 mg/kg group died 7 and 9 days after the last dose, and the remaining animals in the same group were euthanized 9 days after the last dose.

Toxic findings observed were increased white blood cell count and increased levels of hepatic enzymes (ALT, AST, GGT) (≥26 mg/kg groups), reduced body weight gain, decreased platelet count, increased bilirubin, extramedullary hemopoiesis in spleen and liver, decreased bone marrow cell count, hypertrophy of hepatocytes, and increased mitotic figure count (52 mg/kg group).

3.(iii).A.(2).2) Three-month repeat dose study in cynomolgus monkeys

Trastuzumab emtansine (0 [vehicle], 3, 10, 30 mg/kg) was administered intravenously once every 3 weeks for 4 doses in total to cynomolgus monkeys (7/sex/group). Two days after the last dose, 3 each of males and females per group were necropsied and subjected to examination for the repeat dose toxicity of trastuzumab emtansine. At 21 and 42 days after the last dose, 2 each of males and females per group were necropsied to investigate the reversibility.

In this study, all animals remained alive up to the scheduled necropsy date, and did not show any toxic findings in general conditions, body weight, neurological examination, ophthalmological examination, electrocardiogram, or urine analysis. Tests performed 2 days after the last dose showed the following findings: (a) hematology findings included increased neutrophil count and increased fibrinogen (≥10 mg/kg groups), decreased red blood cell parameters (red blood cell count, hemoglobin, hematocrit), decreased platelet count, decreased lymphocyte count, and increased activated partial thromboplastin time (30 mg/kg group); (b) clinical chemistry findings included increased AST, increased globulin, and decreased A/G ratio (≥10 mg/kg groups), increased ALT, increased ALP, and increased triglycerides (30 mg/kg group); and (c) histopathological findings included hypertrophy of hepatic Kupffer cells, vacuolization of hepatocytes, hypertrophy of reticuloendothelial cells in spleen, increased splenic red pulp cell count, depletion of thymic lymphocytes, depletion of lymphocytes in mesenteric lymph nodes, increased mitotic figure count (trastuzumab emtansine groups), axonal degeneration of spinal and sciatic nerves, increased white blood cell count in sinusoidal capillaries of the liver, and multinuclear hepatocytes (≥10 mg/kg groups). The tests performed 42 days after the last dose showed that the toxic findings were reversible except increased ALT, increased globulin, and axonal degeneration.

From these results, HNSTD was determined to be 10 mg/kg.

3.(iii).A.(2).3) Six-month repeat dose study in cynomolgus monkeys

Trastuzumab emtansine (0 [vehicle], 1, 3, 10 mg/kg) was administered intravenously once every 3 weeks for 8 doses in total to cynomolgus monkeys (6 each of males and females per group). At 7 days after the last dose, 3 each of males and females per group were necropsied and subjected to examination for the repeat dose toxicity of trastuzumab emtansine. At 42 days after the last dose, all remaining animals were necropsied to investigate the reversibility.

In this study, all animals remained alive up to the scheduled necropsy date, and did not show any toxic findings in general conditions, body weight, neurological examination, ophthalmological examination, electrocardiogram, blood pressure, urine analysis, necropsy, or organ weight. The tests performed 7 days after the last dose showed the following findings: (a) hematology findings included decreased platelet count, increased monocyte count, and increased fibrinogen (10 mg/kg group); (b) clinical chemistry findings included increased AST and decreased inorganic phosphorus (≥3 mg/kg groups), increased cholesterol and increased creatinine kinase (10 mg/kg group); and (c) histopathological findings included increased mitotic figure count in multiple organs (≥1 mg/kg groups), hypertrophy of epithelial cells and decreased mucous cells in the lacrimal gland, hypertrophy of Kupffer cells and sinusoidal endothelial cells in the liver (≥3 mg/kg group), vacuolization and atrophy of hepatocytes, and apoptosis of prostatic gland and seminal vesicular epithelium (10 mg/kg group). Tests performed 42 days after the last dose showed axonal degeneration of sciatic nerve (≥1 mg/kg groups) along with the persisting decrease in mucous cells in the lacrimal gland, hypertrophy of Kupffer cells and sinusoidal endothelial cells in the liver, atrophy of hepatocytes (≥3 mg/kg groups), and increased mitotic figure count in the gallbladder epithelium (10 mg/kg group), whereas other toxicity findings tended to recover.

Based on the above results, HNSTD was determined to be 10 mg/kg. The exposure levels ($AUC_{147\text{-}168}$) of trastuzumab emtansine at this dose were 1040 μg·day/mL in males and 910 μg·day/mL in females, which were 2.9 and 2.5 times, respectively, the clinical exposure level.* The exposure levels ($AUC_{147\text{-}168}$) of DM1 were 14.9 ng·day/mL in males and 9.31 ng·day/mL in females, which were 2.7 and 1.7 times, respectively, the clinical exposure level.*

*: AUC_{last} in Cycle 1 in Japanese patients receiving trastuzumab emtansine at 3.6 mg/kg every 3 weeks in the Japanese phase II study (Study JO22997)

3.(iii).A.(3) Genotoxicity

In the 6-month repeat dose study in cynomolgus monkeys, bone marrow smear samples were prepared at necropsy 7 days after the last dose, and counted for polychromatic erythrocytes with micronuclei. As a result, no increase in the frequency of micronuclei was observed.

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

Since trastuzumab emtansine is indicated for inoperable or recurrent breast cancer, no carcinogenicity test was conducted.

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

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

In the single intravenous dose study of trastuzumab emtansine in rats, degeneration of testicular seminiferous tubules, cell debris in the ductus epididymis, and necrosis of corpora lutea in the ovary were observed [see "3.(iii).A.(1).1) Single intravenous dose study in rats"].

The applicant explained that trastuzumab emtansine at the recommended clinical dose is unlikely to affect the reproductive system, taking account of the following facts: (1) toxic findings of the reproductive system were observed only in the group receiving the dose of 60 mg/kg, which caused death in some animals; (2) AUC of exposure in the 20 mg/kg group (in which reproductive toxicity was not observed) was 4 times and 2 to 4 times the AUC of exposure at the clinical dose for trastuzumab emtansine and DM1, respectively; and (3) reproductive toxicity was not observed in single or repeat dose toxicity studies in cynomolgus monkeys.

3.(iii).A.(5).2) Study for effects on embryo-fetal development

Studies for effects on embryo-fetal development were not conducted because trastuzumab emtansine is a teratogenic substance and has a risk of fetal toxicity, as demonstrated by the following findings.

- Post-marketing case reports on trastuzumab have shown that administration of the drug during pregnancy is associated with oligohydramnios as well as fatal renal failure and pulmonary hypoplasia of fetuses (see package inserts of Herceptin Intravenous Infusion 60 and Herceptin Intravenous Infusion 150).

- In a reproductive and developmental toxicity study in mice in which maytansine, a DM1-related compound, was administered intraperitoneally, increased frequency of embryo-fetal death, decreased fetal body weight, and abnormalities in fetuses (hydrencephalus, gastroschisis, microphthalmia and anophthalmia, exencephalia, spina bifida, microtia, skeletal abnormalities [ribs, spine]) were observed (*Teratology*. 1978;18:31-48).

- DM1, which is a tubulin polymerization inhibitor, induced a dose-dependent increase in the frequency of micronuclei in a micronucleus test in rats, showing the ability to induce chromosomal abnormality [see "3.(iii).A.(7).5).(c) Micronucleus test in bone marrow cells in rats"].

3.(iii).A.(6) Local tolerance

No local tolerance study was conducted. Instead, in repeat dose toxicity studies, macroscopic observation and histopathological examination of the administered site were performed. In rats and cynomolgus monkeys, macroscopic observation did not show any toxic findings at the administration site, whereas histopathological examination revealed an increase in mitotic figure count in the basal laminae of the epidermis and the appendages. These findings were considered to be due to the inhibitory effect of DM1 on tubulin polymerization, and recovered at the end of the 3-week recovery period.

3.(iii).A.(7) Other toxicity studies

3.(iii).A.(7).1) Tissue cross-reactivity study using normal tissues of humans and cynomolgus monkeys

Cross reactivity of trastuzumab emtansine was investigated by immunohistochemical staining using frozen sections of normal tissues of humans and cynomolgus monkeys. The tissues that showed cross-reactivity with trastuzumab emtansine were identical with those reported to express HER2 (*Oncogene.* 1990;5:953-62).

3.(iii).A.(7).2) Study on hemolysis and hemocompatibility in humans and cynomolgus monkeys

When tested using the whole blood, plasma, and serum of humans and cynomolgus monkeys, trastuzumab emtansine (0 [vehicle], 1.25, 2.5, 5 mg/mL) did not have a hemolytic effect on red blood cells, neither did it have a precipitating or agglutinating effect on the plasma or serum.

3.(iii).A.(7).3) Studies on the mechanism causing decrease in platelet count

As a result of the following investigations on the mechanism causing decreased platelet count [see "3.(iii).A.(1) Single dose toxicity" and "3.(iii).A.(2) Repeat dose toxicity"], neither trastuzumab emtansine nor DM1 directly affected platelet aggregation or platelet activation. However, it was suggested that DM1, upon uptake of trastuzumab emtansine into cells by a HER2-independent and partially Fc-dependent mechanism, causes decreases in megakaryocyte progenitor cell and megakaryocyte counts.

- Trastuzumab emtansine (100 μg/mL), trastuzumab (100 μg/mL), DM1-conjugated anti-CD22 antibody (100 μg/mL) or DM1 (0.01-100μM) was added to platelet samples collected from healthy adult subjects. DM1 at 1 μM (740 ng/mL), the concentration corresponding to approximately 200 times the clinical exposure level of DM1 (C_{max} 3.78 ng/mL), affected neither platelet aggregation nor activation; nor did DM1 have any effect on platelet aggregation or activation induced by factors acting on platelets (collagen, thrombin receptor activating peptide-6 [TRAP6]).

- Trastuzumab emtansine (25 μg/mL), trastuzumab (25 μg/mL), DM1-conjugated anti-glycoprotein D antibody (5B6-DM1) (25 μg/mL), or vehicle was added to human hematopoietic stem cells collected from the bone marrow of healthy adult subjects. Trastuzumab emtansine and 5B6-DM1 caused a marked decrease in megakaryocyte count. In a different study, mature megakaryocytes were treated in a similar manner as described above. Trastuzumab emtansine and 5B6-DM1 also caused a decrease in megakaryocyte count, albeit to a smaller extent as compared with the decrease observed when added to human hematopoietic stem cells.

- Trastuzumab emtansine (25 μg/mL), trastuzumab (25 μg/mL), 5B6-DM1 (25 μg/mL), or vehicle was added to megakaryocytes undergoing differentiation and maturation. The amount of DNA increased from diploid to 128-ploid with differentiation in the samples treated with the vehicle or trastuzumab, whereas no increase in ploidy was observed in the samples treated with trastuzumab emtansine or 5B6-DM1. The effects of trastuzumab emtansine and 5B6-DM1 were considered to be due to the inhibition by DM1 of tubulin polymerization.

- HER2 mRNA or protein was not detected in megakaryocytes or platelets isolated from healthy adult subjects, suggesting that the effect of trastuzumab emtansine on platelets was HER2-independent.

- In the presence of anti-CD32 antibody which inhibits the binding of the Fc region of IgG to FcγRIIb, the amount of trastuzumab emtansine bound to the surface of megakaryocytes or incorporated into the cells was markedly reduced. Thus, it was suggested that the intracellular incorporation of trastuzumab emtansine was partially FcγRIIb-dependent.

- Trastuzumab-MCC-[^{3}H]DM1 was added to megakaryocyte progenitor cells, megakaryocytes undergoing differentiation, and mature megakaryocytes. Radioactivity increased in the acetonitrile-soluble fraction of megakaryocyte progenitor cells and differentiating megakaryocytes, suggesting the release of DM1 in these cells.

3.(iii).A.(7).4) Toxicity studies on free maytansinoids

(a) [redacted].

(b) Single intravenous dose study on trastuzumab emtansine with different free maytansinoid contents in rats

Trastuzumab emtansine (0 [vehicle], 50 mg/kg) containing 2.5%, 3.3%, or 4.8% of free maytansinoids was administered intravenously in a single dose to SD rats (6/sex/group), and animals were monitored for 12 days. In the group treated with trastuzumab emtansine containing 2.5% of free maytansinoids, one animal died and another animal was euthanized, whereas no death or euthanasia occurred in other groups. Other toxicity findings observed were decreased body weight, increased white blood cell count, decreased platelet count, increased levels of hepatic enzymes (ALT, AST, GGT), and increased bilirubin. These toxicity findings were similar irrespective of the free maytansinoid content.

(c) Single intravenous dose study on trastuzumab emtansine with high free maytansinoid content in cynomolgus monkeys

Trastuzumab emtansine (0 [vehicle], 3, 10, 30 mg/kg) containing 5% to 7% of free maytansinoids was administered intravenously in a single dose to cynomolgus monkeys (6 each of males and females per group), and animals were monitored for 3 or 22 days. No death occurred in this study. A transient decrease in body weight was observed in females of the 30 mg/kg group, whereas no toxicity findings were observed in general conditions, food intake, urine analysis, necropsy, or organ weight. In tests performed 3 days after administration, toxicity findings included increased mitotic figure count (hepatic Kupffer cells, etc.) (≥3 mg/kg groups), increased white blood cell count, increased neutrophil count, increased monocyte count, decreased platelet count, increased AST (≥10 mg/kg groups), and increased ALP (30 mg/kg group). Tests performed 22 days after administration showed axonal degeneration and necrosis of sciatic nerve in 1 each of a male and a female in the 30 mg/kg group, but other toxicity findings recovered or tended to recover. The toxicity findings observed in this study were not different from those observed in the single intravenous dose study in cynomolgus monkeys (trastuzumab emtansine containing 2.4% of free maytansinoids was used) [see "3.(iii).A.(1).2) Single intravenous dose study in monkeys"].

3.(iii).A.(7).5) Toxicity studies on DM1

(a) Single intravenous dose study in rats

DM1 (0 [vehicle], 0.07, 0.1, 0.2 mg/kg) was administered intravenously in a single dose to SD rats (10/sex/group), and 5 each of males and females were necropsied 3 days after administration, and remaining animals 22 days after administration. No death occurred. One female in the 0.1 mg/kg group was euthanized 17 days after administration, and one male in the 0.2 mg/kg group 13 days after administration, both because of tail disorders probably caused by the extravascular leakage of the drug solution.

Tests performed 3 days after administration showed the following findings: (a) hematology findings included decreased platelet count and decreased reticulocyte count (≥0.07 mg/kg groups), increased neutrophil count and decreased lymphocyte count (≥0.1 mg/kg groups); (b) clinical chemistry findings included increased ALT and AST (≥0.1 mg/kg groups), increased urea nitrogen, increased cholesterol, decreased total protein, and decreased albumin (0.2 mg/kg group); (c) findings on organ weight included increased liver weight (0.2 mg/kg group); and (d) histopathological findings included hypertrophy and vacuolization of hepatic Kupffer cells, degeneration and necrosis of hepatocytes, degeneration and necrosis of renal tubules, hemorrhage and decreased cell count of bone marrow, increased mitotic figure count in multiple organs (≥0.07 mg/kg groups), necrosis of thymic lymphocytes, necrosis of duodenal lamina propria, necrosis of rectal mucosal epithelium (≥0.1 mg/kg groups), and depletion of splenic lymphocytes (0.2 mg/kg group). Tests performed 22 days after administration showed a tendency of recovery of toxicity findings observed 3 days after administration, except the increased liver weight and increased mitotic figure count in adrenals.

(b) Bacterial reverse mutation assay
A reverse mutation assay was performed using *Salmonella typhimurium* (TA98, TA100, TA1535, TA1537) and *Escherichia coli* (WP2*uvr*A). No increase in the frequency of reverse mutation was observed regardless of the presence or absence of metabolic activation system, from which the applicant determined that DM1 was not mutagenic.

(c) Micronucleus test in bone marrow cells in rats
In a study in which DM1 (0 [vehicle], 0.01, 0.05, 0.1, 0.2 mg/kg) was administered intravenously in a single dose to SD rats (10 males per group), the number of polychromatic red blood cells containing micronuclei increased in a dose-dependent manner, from which the applicant determined that DM1 induced chromosome abnormality.

3.(iii).B. Outline of the review by PMDA
Based on the submitted data and on the following discussion, PMDA has concluded that trastuzumab emtansine may be used clinically.

Neurotoxicity
In the 3-month repeat dose study of trastuzumab emtansine in cynomolgus monkeys, a total of 14 neurotoxicity findings (axonal degeneration of spine, axonal degeneration of sciatic nerve, hypertrophy or hyperplasia of Schwann cells) were observed in the 10 mg/kg group, whereas in the 6-month repeat dose study in cynomolgus monkeys, there was only one neurotoxicity finding observed in the 10 mg/kg group. PMDA asked the applicant to explain the mechanism of the neurotoxicity and the reason for the difference in the incidence of neurotoxicity between the studies.

The applicant responded as follows:
Vinca alkaloids form a stable complex with GTPase of β-tubulin, thereby inhibiting the hydrolysis of GTP, which results in the inhibition of tubulin polymerization. It is considered that this effect causes the loss, and change in the arrangement, of axonal microtubules, inducing the changes of myelinated and unmyelinated nerve fibers, abnormalities in axonal transport, accumulation of neurofilaments in the cell bodies, etc., and at the same time, changes in the myelin sheath consisted of Schwann cells surrounding the axons as the effect secondary to axonal disorders (*Crit Rev Oncol Hematol.* 2012;82:51-77). Since DM1, a component of trastuzumab emtansine, is a derivative of vinca alkaloid maytansine (*Cancer Res.* 1992;52:127-131), the neurotoxicity observed after repeated dose of trastuzumab emtansine is considered to have been induced by the inhibitory effect of DM1 on tubulin polymerization.

The exposure levels (C_{max}, AUC) of trastuzumab emtansine were similar between the 3- and 6-month repeat dose studies above, whereas the exposure level of free DM1 in the 3-month repeat dose study was approximately 2 times that observed in the 6-month repeat dose study. Given that the neurotoxicity observed after administration of trastuzumab emtansine is caused by the inhibitory effect of DM1 on tubulin polymerization, the difference in the exposure level of free DM1 is considered to be the cause for the difference in the incidence of neurotoxicity.

PMDA considers as follows:
PMDA accepted the applicant's response. Although the mechanism of the release of DM1 from trastuzumab emtansine is unclear at present [see "3.(ii).B. Metabolism of trastuzumab emtansine in plasma"], the elucidation of the mechanism will allow the identification of factors that increase the exposure level of free DM1, thereby possibly contributing to reducing adverse drug reactions. Therefore, new information on the mechanism, whenever it becomes available, should be appropriately provided to clinical practice.

4. Clinical data

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

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

4.(i).A.(1) Analytical methods

4.(i).A.(1).1) Measurement of trastuzumab emtansine (genetical recombination) (T-DM1)

[redacted].

4.(i).A.(1).2) Measurement of trastuzumab (genetical recombination)

[redacted]. Method 2 was used in Japanese phase I and phase II studies (Study JO22591 and Study JO22997, respectively) and in foreign phase III studies (Study TDM4370g/Study BO21977, [EMILIA study]).

4.(i).A.(1).3) Measurement of DM1

Concentration of DM1 released from T-DM1 (free DM1) in human plasma was measured by LC-MS/MS.

4.(i).A.(1).4) Measurement of MCC-DM1 and Lys-MCC-DM1

Concentrations of the metabolite generated by the release of DM1 together with the 4-(N-) cyclohexane-1-carboxylate (MCC) linker (MCC-DM1), and of the metabolite generated by the release of MCC-DM1 together with lysine residue (Lys-MCC-DM1) in human plasma were measured by LC-MS/MS.

4.(i).A.(1).5) Measurement of anti-therapeutic antibody (ATA)

[redacted]. Samples in which ATA was detected by either of the above methods were subjected to an absorption test using T-DM1

and trastuzumab to determine whether ATA-positive* or negative.

In the EMILIA study and Study JO22997 which were conducted using the proposed dosage and administration method, the mean serum concentration of T-DM1 was 1.12 to 3.90 and 1.31 to 1.93 μg/mL, respectively, and the mean serum concentration of total trastuzumab was 6.37 to 13.1 and 5.07 to 11.6 μg/mL, respectively, both at the time point of ATA measurement. Taking account of these results, the applicant explained that ATA measurement was unlikely to be affected by T-DM1 or trastuzumab present in the test samples.

*: In both electrochemical luminescence assay and ELISA, the cut point was set at a false positive rate of approximately 5%, in order to minimize the probability of a false-negative judgment.

The applicant explained that the assay system for neutralizing antibody has not been established at present but is under development.

4.(i).A.(1).6) Method of testing for HER2 expression

HER2 expression in tumor tissue was investigated mainly by the fluorescent *in situ* hybridization (FISH) using HER2 FISH pharmDx kit (Dako) or PathVysion HER2 DNA Probe kit (Abbott Molecular), and by immunohistochemical (IHC) staining using HercepTest (Dako).

4.(i).A.(2) Changes in drug substance manufacturing process during development

During the development of the drug substance, the manufacturing process for the drug substance was changed [see "2.A.(1).3).ii) Manufacturing process development (comparability)]. Among the clinical studies submitted in the present application, three formulations were used in the following way: the formulation manufactured by process D/commercial manufacturing process was used in the EMILIA study, foreign phase II studies (Studies TDM4688g and TDM4450g) and Japanese phase I and II studies (Studies JO22591 and JO22997); the formulation by process A in the foreign phase I study (Study TDM3569g) and a foreign phase II study (Study TDM4258g); and the formulation by process B in foreign phase II studies (Studies TDM4258g, TDM4374g, and TDM4450g).

Tests for comparability of quality attributes were performed on the formulations before and after the change in the manufacturing process from formulation manufactured by process A to formulation manufactured by process D/commercial manufacturing process. The pre- and post-change drug substances were considered to be comparable.

4.(ii) Summary of clinical pharmacology studies

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

The pharmacokinetics (PK) of T-DM1 was investigated in patients with HER2-positive breast cancer following the administration of T-DM1 alone.

Patients previously treated with trastuzumab were also included in the following clinical studies, and trastuzumab was detected in the serum of some of these patients prior to the administration of T-DM1. This fact precluded the appropriate evaluation of PK of total trastuzumab following the administration of T-DM1 alone. Therefore, a detailed description of the results of these studies will be omitted.

4.(ii).A.(1) Foreign phase I study (5.3.3.2.1, Study TDM3569g [April 2006 to June 2009])

An open-label, uncontrolled study was conducted to investigate the maximum tolerated dose (MTD), safety, and PK of T-DM1 in 54 patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including trastuzumab.

One treatment cycle consisted of 21 days. T-DM1 was to be intravenously infused every 3 weeks at the doses of 0.3, 0.6, 1.2, 2.4, 3.6, or 4.8 mg/kg, or every week at the doses of 1.2, 1.6, 2.0, 2.4, or 2.9 mg/kg, over 90 ± 10 minutes (the dosing duration could be reduced to 30 ± 10 minutes in the second and subsequent doses when tolerability was confirmed). Serum concentrations of T-DM1 and total trastuzumab and the plasma concentration of free DM1 were measured.

4.(ii).A.(1).1) Every-3-week dosing

PK parameters of T-DM1 in Cycle 1 were as shown in the following table (24 patients included in PK analysis). After administration, T-DM1 was eliminated in a multiphasic manner. V_{ss} was generally constant regardless of dose and similar to the human plasma volume (42.8 mL/kg) (*Pharm Res.* 1993;10:1093-5). In the 2.4, 3.6, and 4.8 mg/kg groups, CL of T-DM1 was lower, and $t_{1/2}$ longer, than in the 0.3, 0.6, and 1.2 mg/kg groups, demonstrating that the PK of T-DM1 was non-linear at low doses but linear at ≥2.4 mg/kg. The applicant explained the reason for these findings as follows: T-DM1 is considered to be eliminated from the body by two routes; one is mediated by the binding with the target antigen and the other is independent of the target antigen. At a high dose of T-DM1, the elimination pathway mediated by the binding to the target antigen became saturated, resulting in decreased CL values, which were almost constant.

In all treatment groups, C_{trough} and C_{peak} in Cycle 1 were similar to those in Cycle 2 and subsequent cycles. Taking account of these findings together with observed $t_{1/2}$ (1.3-4.1 days), the applicant explained that the serum T-DM1 concentration would generally reach a steady state during Cycle 1 when T-DM1 was administered at 3-week intervals.

PK parameters of T-DM1 (Cycle 1)

Dose (mg/kg)	n	C_{max} (μg/mL)	AUC_{inf} (μg·day/mL)	$t_{1/2}$ (day)	CL (mL/day/kg)	V_{ss} (mL/kg)
0.3	3	9.63 ± 1.73	14.5 ± 3.39	1.3 ± 0.2	21.1 ± 4.45	35.7 ± 7.54
0.6	1	13.3	24.5	1.3	24.5	43.8
1.2	1	20.3	42.9	1.3	27.8	51.8
2.4	1	76.3	330	2.2	7.16	30.7
3.6	15	76.2 ± 19.1	300 ± 65.8	3.1 ± 0.7	12.7 ± 3.56	58.4 ± 12.4
4.8	3	130 ± 7.77	673 ± 12.2	4.1 ± 0.7	7.13 ± 0.125	41.2 ± 6.20

Arithmetic mean ± SD, PK parameters were calculated by a model-independent analysis.

In 19 of 24 patients included in PK analysis, trastuzumab had been detected in the serum before T-DM1 administration. Serum concentration of total trastuzumab before T-DM1 administration ranged from 0.06 to 73 μg/mL, which corresponded to 0.1% to 72% of C_{max} of total trastuzumab following the first dose of T-DM1.

Plasma concentration of free DM1 was below the lower limit of quantitation (0.737 ng/mL) at all time points in the 0.3 mg/kg group, while the concentration could be measured at 1 or 2 time points in the 0.6 and 1.2 mg/kg groups. In the 3.6 and 4.8 mg/kg groups, mean C_{max} was 4.57 and 5.66 ng/mL, respectively, mean AUC_{last} was 6.00 and 7.61 ng·day/mL, respectively, and mean AUC_{inf} was 9.11 and 16.0 ng·day/mL, respectively, showing that the exposure level of free DM1 was lower than that of T-DM1. On and after Day 7 in Cycle 1, concentration of free DM1 in the plasma could not be quantitated in all patients of all groups, with no accumulation of free DM1 being observed following the multiple doses of T-DM1. C_{max} of free DM1 tended to increase with dose of T-DM1, while dose-adjusted AUC_{last} tended to be lower in the 0.3, 0.6 and 1.2 mg/kg groups than in the 2.4, 3.6 and 4.8 mg/kg groups. The applicant explained that the observed differences are probably caused by the fewer number of time points where free DM1 is quantifiable in the ≤1.2 mg/kg groups than in the 2.4 and 4.8 mg/kg groups.

4.(ii).A.(1).2) Every week dosing

PK parameters of T-DM1 in Cycle 1 were as shown in the following table. After administration, T-DM1 was eliminated in a multiphasic manner. C_{max} and AUC_{inf} were dose-proportional, whereas CL, $t_{1/2}$, and V_{ss} were constant regardless of the doses. Based on these results, the applicant explained that, following the administration of T-DM1 every week, PK of T-DM1 after the first dose will be linear within the dose range from 1.2 to 2.9 mg/kg.

V_{ss} was similar to human plasma volume. In all groups, C_{trough} and C_{peak} increased with the increase in the number of times of dose up to the third dose in Cycle 1, whereas C_{trough} and C_{peak} remained comparable in Cycle 2 and the subsequent cycles. Based on these results, the applicant explained that serum T-DM1 concentration will reach a steady state during Cycle 1 when T-DM1 is administered every week.

PK parameters of T-DM1 (Cycle 1)

Dose (mg/kg)	n	C_{max} (μg/mL)	AUC_{inf} (μg·day/mL)	$t_{1/2}$ (day)	CL (mL/day/kg)	V_{ss} (mL/kg)
1.2	3	29.6 ± 5.66	76.2 ± 10.4	2.3 ± 0.6	15.9 ± 2.4	47.5 ± 5.97
1.6	3	34.3 ± 4.81	130 ± 39.7	3.4 ± 0.8	13.0 ± 3.4	59.8 ± 16.6
2.0	3	48.0 ± 9.56	175 ± 41.0	3.1 ± 0.3	11.8 ± 2.4	51.0 ± 8.05
2.4	16	54.8 ± 12.6	199 ± 54.5	3.3 ± 1.1	13.1 ± 4.08	55.4 ± 13.0
2.9	3	78.1 ± 33.9	212 ± 39.0	2.9 ± 0.5	14.0 ± 2.6	57.7 ± 2.21

Arithmetic mean ± SD, PK parameters were calculated by a model-independent analysis.

In 13 of 28 patients included in PK analysis, trastuzumab had been detected in the serum before T-DM1 administration. Serum concentration of total trastuzumab before T-DM1 administration ranged from 0.05 to 57.8 μg/mL, which corresponded to 0.1% to 64% of C_{max} of total trastuzumab following the first dose of T-DM1.

Within the dose range investigated, mean C_{max} and mean AUC_{last} of free DM1 were 2.28 to 3.86 ng/mL and 0.74 to 3.24 ng·day/mL, respectively, showing an approximately dose-dependent increase, except in the 2.0 mg/kg group. The exposure level of free DM1 was lower than that of T-DM1. Neither C_{trough} nor C_{peak} of free DM1 increased with the number of doses, which showed that free DM1 did not accumulate following multiple doses of T-DM1 when T-DM1 was administered every week.

4.(ii).A.(2) Japanese phase I study (5.3.3.2.2, Study JO22591 [[redacted] – ongoing (data cut-off, [redacted])])

An open-label, uncontrolled study was conducted to investigate the MTD, safety, and PK of T-DM1 in 10 patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including trastuzumab.

T-DM1 (1.8, 2.4, 3.6 mg/kg) was to be intravenously infused every 3 weeks over 90 ± 10 minutes (the dosing duration could be reduced to 30 ± 10 minutes in the second and subsequent doses when safety was confirmed). Serum concentrations of T-DM1 and total trastuzumab and the plasma concentrations of free DM1 were measured.

PK parameters of T-DM1 in Cycle 1 were as shown in the following table. After the administration, T-DM1 was eliminated in a multiphasic manner. T-DM1 concentrations peaked 30 minutes or 4 hours after the end of treatment, after which it decreased rapidly until 2 days after dosing, then decreased gradually. AUC_{last}, AUC_{inf}, and C_{max} were dose-proportional, while CL, V_{ss}, and $t_{1/2}$ were generally constant regardless of the doses. Based on the results, the applicant explained that the PK of T-DM1 is linear within the dose range between 1.8 and 3.6 mg/kg. In

Cycle 2 and subsequent cycles, C_{trough} and C_{peak} were similar among cycles, and the accumulation ratio calculated from measured C_{peak} values (R_{obs} [C_{peak}]) (mean, 0.945-1.410) was similar to that calculated from the elimination rate constant K_{el} (R_{kel}) (mean, 1.002-1.027).

PK parameters of T-DM1 (Cycle 1)

Dose (mg/kg)	n	C_{max} (μg/mL)	AUC_{last} (μg·day/mL)	AUC_{inf} (μg·day/mL)	$t_{1/2}$ (day)	CL (mL/day/kg)	V_{ss} (mL/kg)
1.8	1	35.3	139	141	2.39	12.9	57.1
2.4	4	43.4 ± 15.2	203 ± 69.9	204 ± 70.5	2.88 ± 0.317	13.4 ± 6.34	67.6 ± 20.3
3.6	5	82.0 ± 10.0	338 ± 36.2	346 ± 41.1	3.74 ± 1.15	10.6 ± 1.26	59.1 ± 6.62

Arithmetic mean ± SD, PK parameters were calculated by a model-independent analysis.

In 5 of 10 patients included in PK analysis, trastuzumab had been detected in the serum before T-DM1 administration. Serum concentration of total trastuzumab before T-DM1 administration ranged from 0.0478 to 31.8 μg/mL, which corresponded to 0.052% to 39% of C_{max} of total trastuzumab following the first dose of T-DM1.

PK parameters of free DM1 were as shown in the following table. Plasma concentrations of free DM1 peaked 30 minutes after the end of treatment, then decreased over time. C_{max}, AUC_{last}, and AUC_{inf} did not increase with dose. In Cycle 2 and subsequent cycles, C_{peak} of free DM1 remained almost constant. The elimination phase could not be evaluated in 1 of 1 patient in the 1.8 mg/kg group, in 1 of 4 patients in the 2.4 mg/kg group, or in 2 of 5 patients in the 3.6 mg/kg group. The applicant explained that PK parameters such as AUC_{inf} and $t_{1/2}$ consequently could not be calculated in these patients.

PK parameters of free DM1 (Cycle 1)

Dose (mg/kg)	n	C_{max} (ng/mL)	t_{max} (day)	AUC_{last} (ng·day/mL)	AUC_{inf} (ng·day/mL)	$t_{1/2}$ (day)
1.8	1	1.48	0.0850	0.247	–	–
2.4	4	4.18 ± 1.11	0.153 ± 0.0867	6.73 ± 5.33	11.7 ± 7.95*	2.59 ± 2.67*
3.6	5	3.41 ± 1.15	0.0814 ± 0.00385	3.49 ± 2.13	8.28 ± 3.63*	3.12 ± 1.28*

Arithmetic mean ± SD, *: n = 3

The relationship between serum concentrations of T-DM1 or total trastuzumab or plasma concentrations of free DM1 and Fridericia-corrected QT interval (QTcF) was investigated. No clear relationship with QTcF was observed for any of them.

4.(ii).A.(3) Foreign phase II study (5.3.5.2.1, Study TMD4258g [redacted] to [redacted])

An open-label, uncontrolled study was conducted to investigate the efficacy and safety of T-DM1 in 112 patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including HER2-targeted therapy.

T-DM1 (3.6 mg/kg) was to be intravenously infused every 3 weeks over 90 ± 10 minutes (the dosing duration could be reduced to 30 ± 10 minutes in the second and subsequent doses when safety was confirmed). Serum concentrations of T-DM1 and total trastuzumab and the plasma concentrations of free DM1 were measured (the table below).

PK parameters of T-DM1 were similar between Cycle 1 and Cycle 4.

In 47 of 108 patients included in PK analysis, trastuzumab had been detected in the serum before T-DM1 administration. Serum concentrations of total trastuzumab before T-DM1 administration ranged from 0.044 to 66.9 μg/mL.

Concentrations of free DM1 in plasma was quantifiable (lower limit of quantitation, 0.737 ng/mL) only at 30 minutes after the end of T-DM1 treatment in most of the patients. C_{max} of free DM1 remained generally unchanged between Cycle 1 and Cycle 4, and the plasma concentrations of free DM1 were ≤17 ng/mL in Cycle1 to Cycle 4.

PK parameters (Cycles 1 and 4)

	Time of measurement	n	C_{max} (μg/mL)	AUC_{last} (μg·day/mL)	AUC_{inf} (μg·day/mL)	$t_{1/2}$ (day)	CL (mL/day/kg)	V_{ss} (mL/kg)
T-DM1	Cycle 1	101	80.9 ± 20.7	450 ± 126	457 ± 129	3.53 ± 0.7	8.51 ± 2.7	28.4 ± 12.9
	Cycle 4	69	68.9 ± 21.8	461 ± 136	499 ± 260	4.43 ± 1.7	8.41 ± 4.3	45.2 ± 43.0
Total trastuzumab	Cycle 1	104	88.0 ± 30.2	753 ± 407	1040 ± 1030	9.18 ± 10.9	5.64 ± 6.3	46.5 ± 58.4
	Cycle 4	71	85.7 ± 24.7	888 ± 294	1330 ± 1040	11.2 ± 6.3	3.54 ± 2.2	46.1 ± 19.9
Free DM1	Cycle 1	105	5.35 ± 2.03*	–	–	–	–	–
	Cycle 4	83	5.89 ± 2.23*	–	–	–	–	–

Arithmetic mean ± SD, *: unit = ng/mL

4.(ii).A.(4) Foreign phase II study (5.3.5.2.2, Study TDM4374g [[redacted] to [redacted]])

An open-label, uncontrolled study was conducted to evaluate the efficacy and safety of T-DM1 in 110 patients with HER2-positive metastatic breast cancer who had previously been treated with chemotherapy with an anthracycline and a taxane antineoplastic drug, capecitabine (Cape), trastuzumab, and lapatinib tosilate hydrate (lapatinib).

T-DM1 (3.6 mg/kg) was to be intravenously infused every 3 weeks over 90 ± 10 minutes (the dosing duration could be reduced to 30 ± 10 minutes in the second and subsequent doses when safety was confirmed). Serum concentrations of T-DM1 and total trastuzumab and the plasma concentrations of free DM1 were measured (the table below).

PK parameters of T-DM1 were similar between Cycle 1 and Cycle 4.

In 57 of 108 patients included in PK analysis, trastuzumab had been detected in the serum before T-DM1 administration. Serum concentrations of total trastuzumab before T-DM1 administration ranged from 0.0503 to 122 μg/mL.

Concentrations of free DM1 in plasma was quantifiable (lower limit of quantitation, 0.737 ng/mL) only at 30 minutes after the end of T-DM1 treatment in most of the patients. C_{max} of free DM1 remained generally unchanged between Cycle 1 and Cycle 4, and plasma concentrations of free DM1 were ≤25 ng/mL in Cycle 1 to Cycle 4.

PK parameters (Cycles 1 and 4)

	Time of measurement	n	C_{max} (μg/mL)	AUC_{last} (μg·day/mL)	AUC_{inf} (μg·day/mL)	$t_{1/2}$ (day)	CL (mL/day/kg)	V_{ss} (mL/kg)
T-DM1	Cycle 1	105	79.5 ± 21.1	475 ± 141	486 ± 141*1	3.96 ± 0.964*1	8.04 ± 2.97*1	31.2 ± 10.9*1
	Cycle 4	82	78.3 ± 25.6	456 ± 162	–	4.33 ± 0.757*2	7.27 ± 2.49*2	39.3 ± 32.8*2
Total trastuzumab	Cycle 1	105	89.9 ± 31.3	837 ± 460	1150 ± 852	9.38 ± 4.90*3	4.55 ± 2.64*3	43.2 ± 16.3*3
	Cycle 4	82	89.2 ± 29.1	700 ± 250	–	9.96 ± 4.81*4	3.26 ± 1.66*4	39.6 ± 13.3*4
Free DM1	Cycle 1	104	5.36 ± 2.56*5	–	–	–	–	–
	Cycle 4	81	5.07 ± 1.92*5	–	–	–	–	–

Arithmetic mean ± SD, *1: n = 102, *2: n = 67, *3: n = 100, *4: n = 61, *5: unit = ng/mL

4.(ii).A.(5) Foreign phase II study (5.3.5.1-2, Study TDM4450g [September 2008 to August 2011])

An open-label, randomized, comparative study was conducted to compare the efficacy and safety of T-DM1 alone and concomitant use of trastuzumab with docetaxel hydrate (DTX) (trastuzumab/DTX group) in 137 patients (67 patients included in PK analysis) with HER2-positive metastatic or recurrent breast cancer who had not previously been treated with chemotherapy except neoadjuvant or adjuvant chemotherapy.

T-DM1 (3.6 mg/kg) was to be intravenously infused every 3 weeks over 90 ± 10 minutes (the dosing duration could be reduced to 30 ± 10 minutes in the second and subsequent doses when safety was confirmed). Serum concentrations of T-DM1 and total trastuzumab and the plasma concentrations of free DM1 were measured (the table below).

PK parameters of T-DM1 were similar between Cycle 1 and Cycle 5.

In 18 of 67 patients included in PK analysis, trastuzumab had been detected in the serum before T-DM1 administration. Serum concentrations of total trastuzumab before T-DM1 administration ranged from 0.0449 to 27.3 μg/mL.

In Cycles 1 and 5, free DM1 concentrations were quantifiable (lower limit of quantitation, 0.737 ng/mL) only at 30 minutes after the end of T-DM1 treatment in most of the patients. Therefore, PK of free DM1 was evaluated based on C_{max} alone. C_{max} remained generally unchanged between Cycle 1 and Cycle 5, and plasma concentrations of free DM1 were ≤17.6 ng/mL in Cycle 1 to Cycle 5.

PK parameters (Cycles 1 and 5)

	Time of measurement	n	C_{max} (μg/mL)	AUC[*1] (μg·day/mL)	$t_{1/2}$ (day)	CL (mL/day/kg)	V_{ss} (mL/kg)
T-DM1	Cycle 1	62	84.2 ± 30.6	495 ± 158[*2]	3.49 ± 0.743[*2]	8.23 ± 3.95[*2]	30.2 ± 21.3[*2]
	Cycle 5	39	79.1 ± 23.7	473 ± 141[*3]	4.22 ± 0.597[*3]	6.67 ± 1.58[*3]	33.6 ± 12.4[*3]
Total trastuzumab	Cycle 1	60	83.3 ± 20.5	700 ± 260[*4]	5.76 ± 1.96[*4]	6.17 ± 4.31[*4]	38.6 ± 11.8[*4]
	Cycle 5	38	108 ± 71.0	788 ± 323	8.34 ± 2.14[*5]	3.05 ± 0.887[*5]	34.8 ± 10.5[*5]
Free DM1	Cycle 1	63	5.11 ± 2.34[*6]	–	–	–	–
	Cycle 5	50	4.71 ± 2.25[*6]	–	–	–	–

Arithmetic mean ± SD, *1: AUC_{inf} in Cycle 1, AUC_{last} in Cycle 5, *2: n = 60, *3: n = 35, *4: n = 51, *5: n = 34, *6: unit = ng/mL

4.(ii).A.(6) Japanese phase II study (5.3.5.2-3, Study JO22997 [[redacted] – ongoing (data cut-off, [redacted])])

An open-label, uncontrolled study was conducted to investigate the efficacy and safety of T-DM1 in 76 patients (32 patients included in PK analysis) with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including trastuzumab.

T-DM1 (3.6 mg/kg) was to be intravenously infused every 3 weeks over 90 ± 10 minutes (the dosing duration could be reduced to 30 ± 10 minutes in the second and subsequent doses when safety was confirmed). Serum concentrations of T-DM1 and total trastuzumab and the plasma concentrations of free DM1, MCC-DM1, and Lys-MCC-DM1 were measured.

PK parameters of T-DM1, total trastuzumab, and free DM1 were as shown in the following table. The mean C_{peak} and the mean C_{trough} of T-DM1 in Cycle 2 and subsequent cycles were within the range between 66.2 and 78.6 μg/mL and between 1.15 and 1.81 μg/mL, respectively, and R_{obs} (C_{peak}) (0.992) was similar to R_{kel} (1.022).

In 22 of 32 patients included in PK analysis, trastuzumab had been detected in the serum before T-DM1 administration. Serum concentrations of total trastuzumab before T-DM1 administration ranged from 0.144 to 22.7 μg/mL.

In Cycle 1, the plasma concentration of free DM1 peaked (mean, 3.79 ng/mL) 30 minutes after the end of treatment, then decreased to below the lower limit of quantitation (0.737 ng/mL) by Day 15 in all patients. In Cycle 2 and subsequent cycles, the mean C_{peak} remained within the range from 2.93 to 4.58 ng/mL, and the maximum plasma concentration of free DM1 up to Cycle 16 was 6.82 ng/mL. C_{trough} of free DM1 was below the lower limit of quantitation (0.737 ng/mL) at all time points.

PK parameters (Cycle 1)

	C_{max} (μg/mL)	AUC_{last} (μg·day/mL)	AUC_{inf} (μg·day/mL)	$t_{1/2}$ (day)	CL (mL/day/kg)	V_{ss} (mL/kg)
T-DM1	78.8 ± 13.9	357 ± 66.6	365 ± 68.4[*1]	3.62 ± 0.782[*1]	10.3 ± 2.30[*1]	54.9 ± 13.9[*1]
Total trastuzumab	81.6 ± 12.4	526 ± 126	657 ± 231[*1]	8.38 ± 3.55[*1]	6.18 ± 2.35[*1]	64.5 ± 16.7[*1]
Free DM1	3.78 ± 0.931[*2]	5.46 ± 6.68[*3]	–	–	–	–

Arithmetic mean ± SD, n = 31, *1: n = 30, *2: unit = ng/mL, *3: unit = ng·day/mL

In Cycle 1, plasma MCC-DM1 concentration peaked (mean, 8.65 ng/mL) 30 minutes after the end of treatment, then decreased to below the lower limit of quantitation (2.93 ng/mL) by Day 8 in all patients. In Cycle 2 and subsequent cycles, mean C_{peak} of MCC-DM1 remained within the range from 5.60 to 10.4 ng/mL, showing no MCC-DM1 accumulation for multiple doses. C_{trough} of MCC-DM1 was below the lower limit of quantitation at all time points, except in 1 patient in whom MCC-DM1 was quantifiable (7.24 ng/mL) before the administration of Cycle 6.

Plasma Lys-MCC-DM1 concentrations were below the lower limit of quantitation (1.10 ng/mL) at all time points, except in 1 patient in whom Lys-MCC-DM1 was quantifiable (1.41 ng/mL) on Day 8 of Cycle 4.

4.(ii).A.(7) Foreign phase II study (5.3.4.2-1, Study TDM4688g [[redacted] to [redacted]])

An open-label, uncontrolled study was conducted to investigate the effect of T-DM1 on QTc intervals in 51 patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including trastuzumab.

T-DM1 (3.6 mg/kg) was to be intravenously infused every 3 weeks over 90 ± 10 minutes (the dosing duration could be reduced to 30 ± 10 minutes in the second and subsequent doses when safety was confirmed).

The mean change in QTcF after dosing of T-DM1 from baseline (ΔQTcF) was, 15 minutes after the end of treatment in Cycle 1, 1.2 msec and showed a tendency to decrease up to Day 8 of the cycle. Then, ΔQTcF increased, reaching 4.7 msec both at 15 minutes and 1 hour after the end of treament in Cycle 3. The upper limit of 95% CI of ΔQTcF was <10 msec at all time points. On the basis of the above results, the applicant explained that T-DM1 was unlikely to affect QTcF intervals.

The relationship between ΔQTcF and serum concentrations of T-DM1 or total trastuzumab or plasma concentrations of free DM1 was investigated using a non-linear mixed effect model. As a result, ΔQTcF tended to increase with the increase in the exposure level of any of these compounds, but the upper limit of 95% CI of ΔQTcF was <10 msec over the entire concentration ranges observed for each of these compounds in Cycle 1 and almost over the entire concentration ranges observed in Cycle 3.

4.(ii).A.(8) Foreign phase III study (5.3.5.1-1, Study TDM4370g [EMILIA study] [February 2009 – ongoing (data cut-off, January 2012)])

An open-label, randomized, comparative study was conducted to compare the efficacy and safety of T-DM1 alone (T-DM1 group) and concomitant use of lapatinib with Cape (lapatinib/Cape group) in 991 patients (350 patients included in PK analysis) with HER2-positive metastatic or recurrent breast cancer who had previously been treated with chemotherapy with a taxane antineoplastic drug and trastuzumab.

T-DM1 (3.6 mg/kg) was to be intravenously infused every 3 weeks over 90 ± 10 minutes (the dosing duration could be reduced to 30 ± 10 minutes in the second and subsequent doses when safety was confirmed). Serum concentrations of T-DM1 and total trastuzumab and the plasma concentrations of free DM1 were measured (the table below).

The time course of serum T-DM1 concentration and the PK parameters were both similar between Cycle 1 and Cycle 4.

In 216 of 316 patients included in PK analysis, trastuzumab had been detected in the serum before T-DM1 administration. The serum concentrations of total trastuzumab before T-DM1 administration ranged from 0.0633 to 124 μg/mL.

In Cycle 1, free DM1 concentrations was quantifiable (lower limit of quantitation, 0.737 ng/mL) only at 30 minutes after the end of T-DM1 treatment in most of the patients. Therefore, PK of free DM1 was evaluated based on C_{max} alone. C_{max} remained generally unchanged between Cycle 1 and Cycle 4, and plasma concentrations of free DM1 were ≤59.7 ng/mL in all cycles.

PK parameters (Cycles 1 and 4)

	Time of measurement	n	C_{max} (μg/mL)	AUC*1 (μg·day/mL)	$t_{1/2}$ (day)	CL (mL/day/kg)	V_{ss} (mL/kg)
T-DM1	Cycle 1	292	83.4 ± 16.5	489 ± 122	3.68 ± 0.886	7.81 ± 2.18	29.5 ± 14.6
	Cycle 4	257	85.0 ± 33.4	475 ± 127	4.19 ± 0.679	7.10 ± 1.89	33.3 ± 11.4
Total trastuzumab	Cycle 1	291	86.3 ± 20.1	816 ± 422	7.80 ± 4.01	5.35 ± 2.33	42.2 ± 15.6
	Cycle 4	256	87.4 ± 30.7	604 ± 166	6.92 ± 2.22	4.68 ± 1.93	41.4 ± 14.5
Free DM1	Cycle 1	287	4.61 ± 1.61*2	–	–	–	–
	Cycle 4	267	5.13 ± 4.09*2	–	–	–	–

Arithmetic mean ± SD, *1: AUC_{inf} in Cycle 1, AUC_{last} in Cycle 4, *2: unit = ng/mL

4.(ii).A.(9) Population pharmacokinetic (PPK) analysis

A population pharmacokinetic (PPK) analysis was conducted using a 2-compartment model with first-order elimination by a nonlinear mixing effect model, based on PK data from 671 patients with breast cancer in 5 studies (9934 time points): a foreign phase I study (Study TDM3569g), foreign phase II studies (Studies TDM4258g, TDM4374g, and TDM4450g), and a foreign phase III study (EMILIA study). As covariates of PK parameters (CL, V_c, Q, V_p) of T-DM1, the following parameters were investigated: body weight, body mass index (BMI), body surface area (BSA), race, sum of longest diameter of target lesions, number of non-target lesions, number of metastases, presence/absence of bone metastases, presence/absence of liver metastases, HER2 ECD concentration, serum trastuzumab concentration at baseline, alanine aminotransferase (ALT), aspartate aminotransferase (AST), serum albumin, alkaline phosphatase (ALP), international normalized ratio (INR), creatinine clearance (CrCL), serum creatinine concentration, region (Asia, non-Asia), and status of systemic therapy.

The applicant explained the results of PPK analysis as follows:

- As statistically significant covariates of the PK parameters of T-DM1, body weight, HER2 ECD concentration, serum albumin, sum of longest diameter of target lesions, serum

trastuzumab concentration at baseline, and AST were selected for CL, and body weight for V_c. CL increased in patients with increased body weight, HER2 ECD concentration, sum of longest diameter of target lesions, or AST, and decreased in patients with increased serum albumin or serum trastuzumab concentration at baseline. V_c increased in patients with increased body weight.

- CL of average patients (body weight, 70 kg; HER2 ECD concentration, 25 ng/mL; serum albumin, 41 g/L; sum of longest diameter of target lesions, 9 cm; serum trastuzumab concentration at baseline, 0 μg/mL; AST, 27 U/L) was estimated to be 0.676 L/day (inter-individual variability, 19.11%), and V_c of average patients (body weight, 70 kg) to be 3.127 L (inter-individual variability, 11.66%). Q and V_p in the final model were estimated to be 1.534 L/day (inter-individual variability, 180.8%) and 0.660 L (inter-individual variability, 74.5%), respectively.

- Effects of factors selected as statistically significant covariates for PK parameters of T-DM1 (body weight, HER2 ECD concentration, serum albumin, sum of longest diameter of target lesions, serum trastuzumab concentration at baseline, AST) on the exposure level of T-DM1 (AUC, C_{max}, C_{trough}) at steady state were investigated. As a result, body weight was selected as the covariate that had the greatest effect on AUC and C_{max} of T-DM1 at steady state.

Based on the above results, the applicant explained as follows:
The results suggested that body weight had the greatest effect on the exposure level of T-DM1 as a covariate, but the effect was only minor and clinically insignificant. Therefore, it is unnecessary to adjust the dose of T-DM1 depending on the above covariates (body weight, HER2 ECD concentration, serum albumin, sum of longest diameter of target lesions, serum trastuzumab concentration at baseline, AST).

4.(ii).A.(10) Effect of ATA on PK of T-DM1
Production of ATA was investigated in a Japanese phase I study ([a] Study JO22591, 10 patients), a foreign phase I study ([b] Study TDM3569g, 48 patients), a Japanese phase II study ([c] Study JO22997, 73 patients), foreign phase II studies ([d] Study TDM4258g, 108 patients, [e] Study TDM4374g, 108 patients, [f] Study TDM4688g, 47 patients, [g] Study TDM4450g, 65 patients), and a foreign phase III study ([h] EMILLIA study, 460 patients). ATA was measured by electrochemical luminescence assay in Studies TDM3569g and TDM4258g and by ELISA in other studies.

The applicant explained the result of the investigation as follows:
ATA was assessed as positive in 44 of 908 patients (4.8%) before T-DM1 administration and in 47 of 919 patients (5.1%) after administration in the above clinical studies. Thirty of 44 patients who were assessed as ATA-positive before T-DM1 administration were considered to be false-positive because they were negative after the administration. Comparison of PK (AUC_{inf} and C_{max} in Cycle 1) between ATA-positive patients and ATA-negative patients in 5 foreign studies did not show any clear difference. The effect of ATA on the PK of T-DM1 could not be clarified from the results of Japanese clinical studies because of the limited number of ATA-positive patients, 1 and 2 patients in Studies JO22591 and JO22997, respectively.

4.(ii).A.(11) Pharmacokinetic interactions
It is reported that when T-DM1 and pertuzumab are co-administered, pertuzumab does not affect the PK of T-DM1, nor does T-DM1 affect the PK of pertuzumab (*Curr Drug Metab.* 2012;13:911-22).

The applicant explained the pharmacokinetic interactions of T-DM1 as follows:

Nonclinical studies have shown that DM1, a component of T-DM1, is metabolized mainly by CYP3A4 and serves as a substrate for P-gp [see "3.(ii).A.(6) Pharmacokinetic interactions"]. However, no clinical studies were conducted aimed at investigating the pharmacokinetic interactions of T-DM1. Therefore, effects of combination of T-DM1 with CYP3A4 inhibitors, CYP3A4 inducers, or P-gp inhibitors on the PK of T-DM1, total trastuzumab, and free DM1 after T-DM1 administration were investigated based on the PK data obtained in the foreign phase III study (EMILIA study). Results showed that PK parameters of T-DM1, total trastuzumab, and free DM1 were similar regardless of the type of drugs concomitantly administered (CYP3A4 inhibitors, CYP3A4 inducers, or P-gp inhibitors). These results suggested that the PK of T-DM1, total trastuzumab, and free DM1 after T-DM1 administration were unaffected by concomitant medication with CYP3A4 inhibitors, CYP3A4 inducers, and P-gp inhibitors. However, taking account of the fact that no clinical studies were conducted aimed at investigating the pharmacokinetic interactions of T-DM1, the information of *in vitro* studies that free DM1 served as the substrate for CYP3A4 and P-gp should be provided appropriately via the package insert.

4.(ii).A.(12) Effect of impaired hepatic or renal function on PK of T-DM1

No clinical studies were conducted in patients with hepatic or renal impairment to investigate the PK of T-DM1. However, the applicant explained that, at present, there is no need for adjusting the dose of T-DM1 in patients with hepatic or renal impairment for the following reasons.

- In PPK analysis, CrCL, an index for renal function, was not selected as a covariate for PK parameters of T-DM1 (CL, V_c, Q, V_p) [see "4.(ii).A.(9) Population pharmacokinetic (PPK) analysis"].

- Bayesian estimates of CL and V_c of T-DM1 based on the final model of the above PPK analysis were classified by the severity of renal impairment (normal, CrCL ≥90 mL/min; mild, CrCL ≥60 mL/min and <90 mL/min; moderate, CrCL ≥30 mL/min and <60 mL/min; severe, CrCL ≥15 mL/min and <30 mL/min). Results showed that CL and V_c in patients with normal renal functions (361 patients), mild impairment (254 patients), moderate impairment (53 patients), and severe impairment (1 patient) were distributed within similar ranges, which suggested that the PK of T-DM1 was not affected by renal function.

- In PPK analysis, serum albumin and AST, indices for hepatic function, were selected as statistically significant covariates for CL of T-DM1. However, sensitivity analysis showed that the extent of the effect of these covariates on AUC, C_{max}, and C_{trough} at steady state were <10%, <1%, and <31%, respectively, which suggested that serum albumin and AST had only minor effects on the PK of T-DM1.

Since patients with hepatic impairment were excluded from clinical studies of T-DM1, results of PPK analysis were obtained from a limited range of patients. In addition, DM1 is excreted mainly into feces via bile in rats [see "3.(ii).A.(5) Excretion"]. Therefore, a clinical pharmacology study (Study BO25499) is being conducted to investigate the effect of impaired hepatic function on the PK and safety of T-DM1.

4.(ii).A.(13) Effect of serum trastuzumab concentration or HER2 ECD concentration on PK of T-DM1

Trastuzumab present in the blood at baseline could possibly affect the elimination of T-DM1 via the target antigen (HER2)-dependent pathway. In addition, it was reported that the HER2 ECD concentration at baseline is a covariate affecting the PK of trastuzumab (*Cancer Chemother Pharmacol.* 2005;56:361-9), suggesting that HER2 ECD concentration could affect the PK of T-DM1 as well. Therefore, the relationship between the exposure level of T-DM1 and serum trastuzumab concentration or HER2 ECD concentration at baseline was investigated.

The relationship between the exposure level of T-DM1 and serum trastuzumab concentration at baseline was investigated in each of Studies TDM4258g, TDM4374g, TDM4450g, TDM4688g, and the EMILIA study. Also, the relationship between the exposure level of T-DM1 and HER2 ECD concentration at baseline was investigated in each of Studies JO22997, TDM4258g, TDM4374g, TDM4450g, and the EMILIA study. As a result, no clear relationship was observed between the exposure level of T-DM1 and serum trastuzumab concentration or HER2 ECD concentration at baseline in any of the studies.

As a result of PPK analysis, serum trastuzumab concentration and HER2 ECD concentration at baseline were selected as covariates affecting the PK of T-DM1, but it was shown that the effect of these covariates on the PK of T-DM1 was minimal [see "4.(ii).A.(9) Population pharmacokinetic (PPK) analysis"].

Based on the above, the applicant explained that neither serum trastuzumab concentration nor HER2 ECD concentration at baseline is likely to have a clear effect on the PK of T-DM1.

4.(ii).A.(14) Relationship between exposure level and efficacy or safety

(a) Relationship between exposure level and efficacy

In the EMILIA study, the relationship between the efficacy and the exposure level of T-DM1, total trastuzumab, or free DM1 following T-DM1 administration was investigated. Patients were classified into subgroups by median AUC_{last} of T-DM1, median AUC_{last} of total trastuzumab, or median C_{max} of free DM1 in Cycle 1, and subjected to comparison of overall survival (OS, obtained from the results of an interim analysis performed at the time of the final analysis of progression-free survival [PFS]) and PFS by Kaplan-Meier curves. No clear relationship was observed between AUC_{last} of T-DM1, AUC_{last} of total trastuzumab, or C_{max} of free DM1 in Cycle 1 and OS or PFS. In addition, the exposure levels of T-DM1, total trastuzumab, and free DM1 were similar between patients who responded to the treatment and those who did not.

In Studies TDM4258g and TDM4374g, no clear relationship was observed either between AUC_{last} of T-DM1, AUC_{last} of total trastuzumab, or C_{max} of free DM1 in Cycle 1 and response.

In Study JO22997, the relationship between AUC_{last} of T-DM1, AUC_{last} of total trastuzumab, or C_{max} of free DM1 following T-DM1 administration in Cycle 1 and the best overall response was investigated. The exposure levels of T-DM1, total trastuzumab, and free DM1 tended to be higher in patients with partial response (PR) than in patients with progressive disease (PD), whereas the exposure levels were similar among patients with PR, stable disease (SD), and PD.

(b) Relationship between exposure level and safety

Based on the results of 5 foreign clinical studies in 618 patients with HER2-positive breast cancer, the relationship between safety and the exposure level of T-DM1, total trastuzumab, or free DM1 following T-DM1 administration was investigated.

No clear relationship was observed between C_{max} or AUC_{last} of T-DM1, AUC_{last} of total trastuzumab, or C_{max} of free DM1 in Cycle 1 and the incidence of Grade 3 or 4 thrombocytopenia or hepatotoxicity. Patients were classified into subgroups by quartile points of AUC_{last} of T-DM1 or total trastuzumab or C_{max} of free DM1 in Cycle 1, and subjected to comparison of time course of laboratory values (ALT, AST, total bilirubin, platelet count) in Cycles 1 to 16. Because of the large inter-individual variability of laboratory values, no clear relationship was observed between the exposure level of T-DM1, total trastuzumab, or free DM1 and the time course of laboratory values.

In Study JO22997, patients were classified into subgroups by quartile points of AUC_{inf} of T-DM1 in Cycle 1 and subjected to comparison of the time course of laboratory values (platelet count,

AST, ALT) in Cycles 1 to 16. As were the cases with the foreign studies, no clear relationship was observed between the exposure level of T-DM1 and the time course of laboratory values because of the large inter-individual variability of laboratory values.

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

4.(ii).B.(1) PK in Japanese and foreign patients

The applicant explained that there were no clear differences in the PK of T-DM1 between Japanese and foreign patients based on the following results.

- PK data of T-DM1, total trastuzumab, and free DM1 obtained from the 3.6 mg/kg group were compared between the Japanese phase I study (Study JO22591) and the foreign phase I study (Study TDM3569g) in which blood samples were collected under the same conditions as in Study JO22591. As a result, C_{peak} and C_{trough} of T-DM1, total trastuzumab, and free DM1 in Cycles 1 to 10 showed similar changes over time in the 2 studies. In addition, the distributions of AUC_{inf} and C_{max} of T-DM1 and total trastuzumab, and C_{max} of free DM1 in Cycle 1 were within similar ranges between the 2 studies.

- PK data of T-DM1, total trastuzumab, and free DM1 obtained from the 3.6 mg/kg group were compared between the Japanese phase II study (Study JO22997) and the foreign phase II studies (Studies TDM4258g and TDM4374g) in which blood samples were collected under the same conditions as in Study JO22997. As a result, the mean AUC_{inf} of T-DM1 and total trastuzumab in Cycle 1 tended to be lower in Study JO22997 compared with Studies TDM4258g and TDM4374g, but the distributions of AUC_{inf} and C_{max} of T-DM1, and total trastuzumab and C_{max} of free DM1 in Cycle 1 were generally within similar ranges between the 2 studies.

- Using PK data obtained from Japanese clinical studies (Studies JO22591 and JO22997) and foreign clinical studies (Studies TDM3569g, TDM4258g, TDM4374g, and TDM4450g, and EMILIA study), PK parameters of T-DM1 were estimated based on the final model of the above PPK analysis to compare CL and V_c between Japanese and foreign patients. Since body weight was suggested as the covariate with the greatest effect on the PK of T-DM1, the comparison was made using CL and V_c standardized for body weight. As a result, the distributions of these PK parameters were similar between Japanese and foreign patients. Using these PK parameters, AUC, C_{max}, and C_{min} of T-DM1 at steady state following multiple doses of T-DM1 (3.6 mg/kg) every 3 weeks were estimated. No clear differences were noted in the exposure level of T-DM1 between Japanese and foreign patients.

PMDA accepted the applicant's explanation.

4.(ii).B.(2) T-DM1 administration in patients with hepatic impairment

The applicant explained that it is unnecessary to adjust the dose of T-DM1 in patients with hepatic impairment [see "4.(ii).A.(12) Effect of impaired hepatic and renal function on PK of T-DM1"].

Based on the submitted data, PMDA considers that the applicant's explanation is generally acceptable that it is unnecessary at the current moment to adjust the dose of T-DM1 in patients with hepatic impairment. However, information on the ongoing Study BO25499 should be provided to clinical practice in an appropriate manner as soon as results become available.

4.(iii) Summary of clinical efficacy and safety

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

As the efficacy and safety evaluation data, the results from a total of 6 studies including 1 each of phase I and phase II studies conducted in Japan and 1 phase I study, 2 phase II studies, and 1 phase III study conducted in foreign countries were submitted. As the reference data, the results from a total of 3 studies including 1 phase I/II study and 2 phase II studies conducted in foreign countries were submitted.

List of clinical studies on efficacy and safety

Data category	Region	Study number	Phase	Patient population	No. of enrollment	Dosage regimen	Primary endpoints
Evaluation	Japan	JO22591	I	Patients with HER2-positive metastatic or recurrent breast cancer previously treated with chemotherapy	10	1.8, 2.4, or 3.6 mg/kg, every 3 weeks	Safety PK
		JO22997	II	Patients with HER2-positive metastatic or recurrent breast cancer previously treated with chemotherapy	76	3.6 mg/kg every 3 weeks	Efficacy
	Foreign	TDM3569g	I	Patients with HER2-positive metastatic or recurrent breast cancer previously treated with chemotherapy	54	(a) 0.3, 0.6, 1.2, 2.4, 3.6, or 4.8 mg/kg every 3 weeks (b) 1.2, 1.6, 2.0, 2.4, or 2.9 mg/kg every week	Safety PK
		TDM4258g	II	Patients with HER2-positive metastatic or recurrent breast cancer previously treated with chemotherapy	112	3.6 mg/kg every 3 weeks	Efficacy
		TDM4374g	II	Patients with HER2-positive metastatic breast cancer previously treated with chemotherapy	110	3.6 mg/kg every 3 weeks	Efficacy
		TDM4370g/ BO21977 (EMILIA)	III	Patients with HER2-positive metastatic or recurrent breast cancer previously treated with chemotherapy with a taxane antineoplastic drug and trastuzumab	991 (a) 495 (b) 496	(a) T-DM1 group: T-DM1 (3.6 mg/kg) every 3 weeks (b) Lap/Cape group: Lap (1250 mg/day) administered orally for 3 weeks and Cape (2000 mg/m^2/day) administered orally for 2 weeks followed by 1-week withdrawal	Efficacy Safety
Reference	Foreign	TDM4373g/ BO22495	I/II	Patients with HER2-positive metastatic or recurrent breast cancer treated or untreated with chemotherapy	67	T-DM1 (2.4, 3.0, 3.6 mg/kg) was administered every 3 weeks. Pertuzumab was administered 840 mg as an initial dose on Day 1 of Cycle 1, and 420 mg in subsequent cycles every 3 weeks.	Safety
		TDM4450g/ BO21976	II	Patients with HER2-positive metastatic or recurrent breast cancer untreated with chemotherapy	137 (a) 67 (b) 70	(a) T-DM1 group: T-DM1 (3.6 mg/kg) every 3 weeks (b) Trastuzumab/DTX group: Trastuzumab (8 mg/kg) and DTX (75 or 100 mg/m^2) were administered on Day 1 of Cycle 1, and trastuzumab (6 mg/kg) and DTX (75-100 mg/m^2) were administered on Day 1 of subsequent cycles every 3 weeks.	Safety
		TDM4688g	II	Patients with HER2-positive metastatic or recurrent breast cancer previously treated with chemotherapy	51	3.6 mg/kg every 3 weeks	Safety

Lap: Lapatinib tosilate hydrate, Cape: Capecitabine, Trastuzumab: Trastuzumab (genetical recombination), DTX: Docetaxel hydrate, PK: Pharmacokinetics

The outline of each clinical study was as described below.

Major adverse events other than deaths reported in each clinical study are described in "4.(iv) Adverse events, etc. observed in clinical studies," and PK data, etc. in "4.(i) Summary of biopharmaceutic studies and associated analytical methods" and in "4.(ii) Summary of clinical pharmacology studies."

Evaluation data

(1) Japanese clinical studies

1) Japanese phase I study (5.3.3.2.2, Study JO22591 [– ongoing (safety data cut-off, ,)])

An open-label, uncontrolled study was conducted to evaluate the safety, tolerability, and PK following intravenous administration of T-DM1 in patients with HER2-positive, metastatic or recurrent breast cancer who had progressed after chemotherapy including trastuzumab (target sample size; ≥1 patient in cohort 1 [1.8 mg/kg every 3 weeks], ≥3 patients in cohort 2 [2.4 mg/kg every 3 weeks], ≥3 patients in cohort 3 [3.6 mg/kg every 3 weeks]) in 3 medical institutions in Japan.

T-DM1 (1.8, 2.4, 3.6 mg/kg) was to be administered intravenously every 3 weeks.

All of the 10 patients enrolled in the study (1 patient in cohort 1, 4 patients in cohort 2, 5 patients in cohort 3) received T-DM1 and were included in the safety analysis.

During the period of dose-limiting toxicity (DLT) evaluation (Cycle 1), 1 patient experienced an adverse event that was considered as DLT. The probability of DLT rate was estimated by the continual reassessment method (CRM) at the time point when the number of patients for the DLT evaluation reached 10. Regardless of the results from patients enrolled thereafter, the dose level at which the probability of DLT rate was closest to 25% (the pre-defined target) was 3.6 mg/kg. Therefore, MTD was estimated to be 3.6 mg/kg.

As regards safety, no death occurred up to 28 days after the last dose of the study drug.

2) Japanese phase II study (5.3.5.2.3, Study JO22997 [, – ongoing (safety data cut-off, ,)])

An open-label, uncontrolled study was conducted to evaluate the efficacy, safety, and PK of T-DM1 in patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including trastuzumab (target sample size, 70) in 29 medical institutions in Japan.

T-DM1 (3.6 mg/kg) was to be administered intravenously every 3 weeks.

Of 76 patients enrolled in the study, 3 patients did not receive T-DM1, and the remaining 73 patients were included in the efficacy and safety analyses.

The best overall response and the response rate, the primary efficacy endpoints assessed by the independent review facility (IRF), were as shown in the following table. The lower limit of the 90% CI of the response rate estimated by the Clopper-Pearson method exceeded the pre-defined threshold of 20%.

Best overall response and response rate (RECIST, efficacy analysis population)

	Number of patients (%) N = 73	
	IRF assessment	Investigator's assessment
Best overall response		
Complete response (CR)	0	0
Partial response (PR)	28 (38.4)	21 (28.8)
Stable disease (SD)	20 (27.4)	33 (45.2)
Progressive disease (PD)	22 (30.1)	16 (21.9)
Unevaluable	3 (4.1)	3 (4.1)
Response (CR + PR) (response rate [90% CI], %)	28 (38.4 [28.8, 48.6])	21 (28.8 [20.2, 38.7])

As regards safety, no death occurred up to 28 days after the last dose of the study drug.

(2) Foreign clinical studies

1) Foreign phase I study (5.3.3.2.1, Study TDM3569g [April 2006 to June 2009])

An open-label, uncontrolled study was conducted to evaluate the safety, tolerability, and PK of T-DM1 in patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including trastuzumab (target sample size, 50-60) in 4 medical institutions overseas.

In each of the 21-day treatment cycles, T-DM1 was administered intravenously every 3 weeks at the doses of 0.3, 0.6, 1.2, 2.4, 3.6, or 4.8 mg/kg, or every week at the doses of 1.2, 1.6, 2.0, 2.4, or 2.9 mg/kg.

Of 54 patients enrolled in the study, 52 patients were included in the safety analysis. Excluded were 2 patients who did not receive T-DM1, one because of abnormal hepatic function due to disease progression and the other because of consent withdrawal.

MTD in every-3-week regimen and every week resimen was 3.6 mg/kg and 2.4 mg/kg, respectively.

As regards safety, no death occurred up to 30 days after the last dose of the study drug.

2) Foreign phase II study (5.3.5.2.1, Study TDM4258g [[redacted] to [redacted]])

An open-label, uncontrolled study was conducted to evaluate the efficacy, safety, and tolerability of T-DM1 in patients with HER2-positive metastatic or recurrent breast cancer who had progressed after chemotherapy including HER2-targeted therapy (target sample size, 100) in 32 medical institutions overseas.

T-DM1 (3.6 mg/kg) was to be administered intravenously every 3 weeks.

All of 112 patients enrolled in the study received T-DM1 and were included in the safety analysis. Efficacy could be evaluated in 109 of the 112 patients at the time point of the primary analysis (after the observation period of approximately 6 months) and in 108 patients at the time point of the final analysis.

The best overall response and the response rate, which were the primary efficacy endpoints assessed by IRF, were as shown in the following table. The lower limit of the 95% CI of the response rate estimated by the Blyth-Still-Casella method exceeded the pre-defined threshold of 14%.

Best overall response and response rate (RECIST, efficacy analysis population)

	Number of patients (%) N = 108	
	IRF assessment	Investigator's assessment
Best overall response		
Complete response (CR)	0	4 (3.7)
Partial response (PR)	29 (26.9)	38 (35.2)
Stable disease (SD)	55 (50.9)	44 (40.7)
Progressive disease (PD)	22 (20.4)	22 (20.4)
Unevaluable	1 (0.9)	0
Missing data	1 (0.9)	0
Response (CR + PR) (response rate [95% CI], %)	29 (26.9 [19.2, 35.8])	42 (38.9 [29.7, 48.5])

As regards safety, 3 patients died within 30 days after the last dose of the study drug. Of these fatal cases, 2 cases were due to disease progression, whereas the remaining 1 case was due to respiratory failure and its causal relationship with the study drug was ruled out.

3) Foreign phase II study (5.3.5.2.2, Study TDM4374g [[redacted] to [redacted]])

An open-label, uncontrolled study was conducted to evaluate the efficacy, safety, and tolerability of T-DM1 in patients with HER2-positive metastatic breast cancer who had previously been treated with chemotherapy with an anthracycline and a taxane antineoplastic drug, Cape, trastuzumab, and lapatinib (target sample size, 100) in 44 medical institutions overseas.
T-DM1 (3.6 mg/kg) was to be administered intravenously every 3 weeks.

All of the 110 patients enrolled in the study received T-DM1 and were included in the efficacy and safety analyses.

The best overall response and the response rate, which were the primary efficacy endpoints assessed by IRF, were as shown in the following table. The lower limit of the 95% CI of the response rate estimated by the Blyth-Still-Casella method exceeded the pre-defined threshold of 14%.

Best overall response and response rate (RECIST, efficacy analysis population)

	Number of patients (%) N = 110	
	IRF assessment	Investigator's assessment
Best overall response		
Complete response (CR)	0	2 (1.8)
Partial response (PR)	36 (32.7)	34 (30.9)
Stable disease (SD)	51 (46.4)	56 (50.9)
Progressive disease (PD)	20 (18.2)	16 (14.5)
Unevaluable	2 (1.8)	1 (0.9)
Missing data	1 (0.9)	1 (0.9)
Response (CR + PR) (response rate [95% CI], %)	36 (32.7 [24.1, 42.1])	36 (32.7 [24.1, 42.1])

As regards safety, 1 patient died within 30 days after the last dose of the study drug. The death was caused by hepatic function abnormal, for which a causal relationship with the study drug could not be ruled out.

4) Foreign phase III study (5.3.5.1.1, Study TDM4370g/BO21977 [EMILIA] [February 2009 – ongoing (data cut-off, January 2012)])

An open-label, randomized, comparative study was conducted to compare the efficacy and safety between the T-DM1 group and the lapatinib/Cape group in patients with HER2-positive metastatic or recurrent breast cancer who had previously been treated with chemotherapy with a

taxane antineoplastic drug and trastuzumab (target sample size, 980) in 213 medical institutions overseas.

In the T-DM1 group, T-DM1 (3.6 mg/kg) was to be administered intravenously every 3 weeks. In the lapatinib/Cape group, lapatinib (1250 mg/day) was to be administered orally for 3 weeks, and Cape (2000 mg/m^2/day) orally for 2 weeks, followed by 1-week withdrawal. The treatment was allowed to continue until progression of disease or intolerable adverse event.

All of the 991 patients enrolled in the study (496 patients in the lapatinib/Cape group, 495 patients in the T-DM1 group) were included in the intent-to-treatment (ITT) population for efficacy analysis. Of patients in the ITT population, 978 patients who received at least one dose of the study drug (490 patients in the T-DM1 group, 488 patients in the lapatinib/Cape group) were included in the safety analysis.

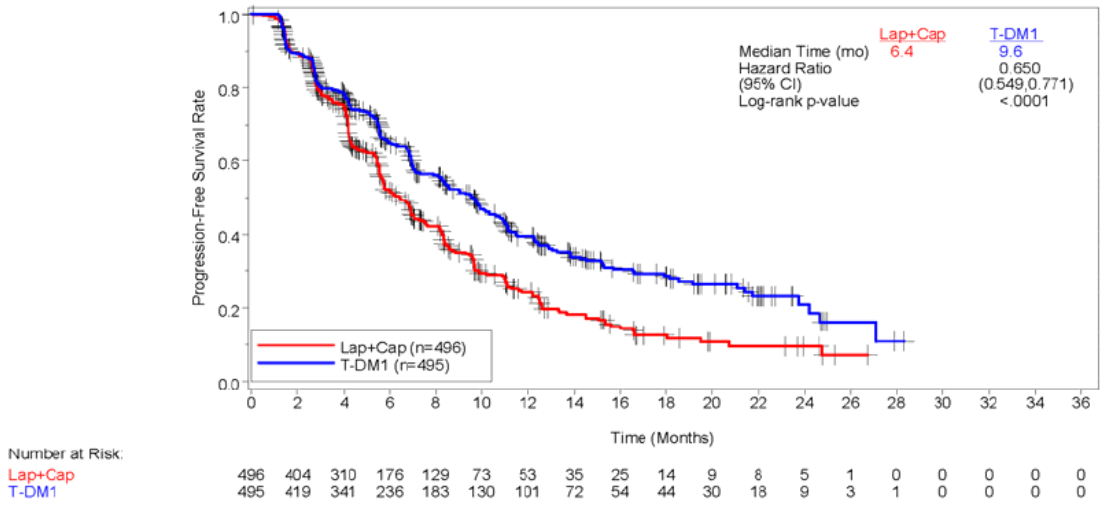
At the initiation of the study, PFS (IRF assessment) was defined as the primary endpoint, and OS as the secondary endpoint. As regards the target number of the PFS events, assuming that the hazard ratio of PFS (IRF assessment) in the T-DM1 group to that of the lapatinib/Cape group is 0.75 (corresponding to median 6.2 months in the lapatinib/Cape group and 8.3 months in the T-DM1 group), approximately 508 events were required to ensure the statistical power of 90% at a two-sided significance level of 5%. Therefore, the target sample size in the study was set at 580 patients. However, based on the discussion with the US Food and Drug Administration, the protocol was amended in October 2010 to handle OS, which had been defined as the secondary endpoint in the previous protocol, as a primary endpoint. Pursuant to this revision, assuming that the hazard ratio of OS in the T-DM1 group to that of the lapatinib/Cape group is 0.8 (corresponding to median 17.2 months in the lapatinib/Cape group and 21.5 months in the T-DM1 group), approximately 632 events were required to ensure the statistical power of 80% at a two-sided significance level of 5%. Therefore, the target sample size in the study was changed to 980 patients. OS was evaluated at an interim analysis at the time point of the final analysis of PFS. In order to control the type I error in the entire study at a two-sided nominal level of 5%, the protocol specified in advance that OS hypothesis be tested using a hierarchical hypothesis structure on a confirmatory basis only if the T-DM1 group is shown to be statistically superior in PFS, and that Lan-DeMets alpha spending function with an O'Brien-Fleming boundary be used in deriving the significance level in the interim analysis.

As regards efficacy, results of the final analysis of PFS (IRF assessment) and its Kaplan-Meier curve were as shown in the following table and figure.

Results of final analysis of PFS (ITT population, IRF assessment, data cut-off date of January 2012)

	T-DM1 group	Lapatinib/Cape group
Number of patients	495	496
Number of events (%)	265 (53.5)	304 (61.3)
Median [95% CI] (months)	9.6 [8.25, 10.64]	6.4 [5.68, 7.06]
Hazard ratio [95% CI] *1	0.650 [0.549, 0.771]	
P value (two-sided)*2	< 0.0001	

*1: The Cox proportional hazard model stratified by number of chemotherapy regimens (0-1 vs. >1), presence or absence of visceral metastasis, world region (US, Western Europe, other), *2: Log-rank test stratified by the number of chemotherapy regimens (0-1 vs. >1), presence or absence of visceral metastasis, world region (US, Western Europe, other), two-sided significance level of 5%



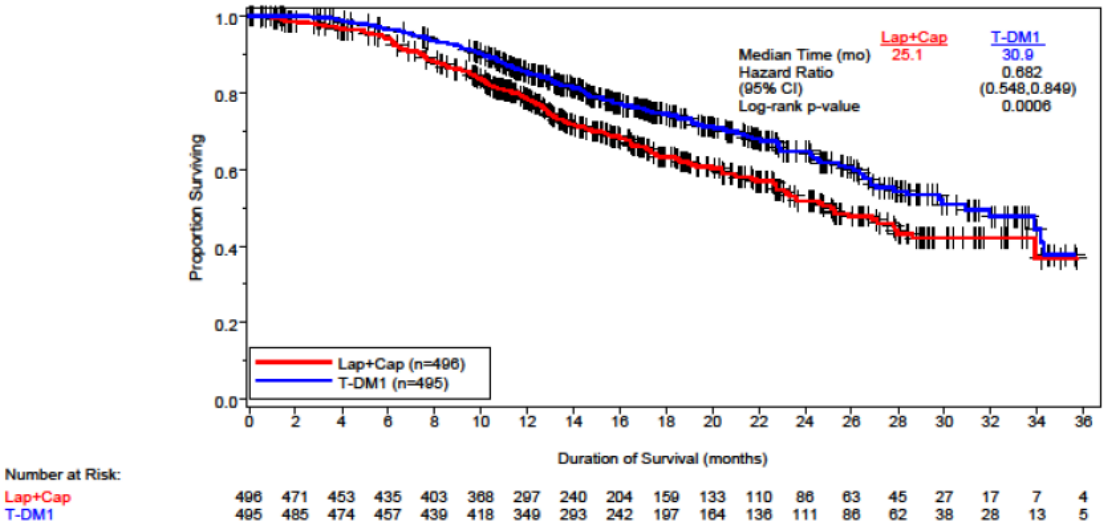
Kaplan-Meier curve of PFS (ITT population, IRF assessment, data cut-off date of January 2012)

Since the above results demonstrated the superiority of T-DM1 in PFS (IRF assessment), the interim analysis of OS was performed according to the protocol specified in advance using the hierarchical hypothesis structure. As a result, the hazard ratio [95% CI] was 0.621 [0.475, 0.813] and the median OS [95% CI] was not estimable [26.32 months, non-estimable] in the T-DM1 group and 23.3 months [20.93 months, non-estimable] in the lapatinib/Cape group (P = 0.0005, stratified long-rank test), failing to meet the criteria for efficacy stopping boundary (two-sided significance level 0.0003). However, since a positive trend in OS improvement was observed, by taking account of the discussion with foreign regulatory agencies, it was decided from ethical considerations for patients to perform an additional interim analysis when at least 50% of the target number of 632 events occurred. Results of the additional interim analysis of OS and its Kaplan-Meier curve were as shown in the following table and figure, respectively, fulfilling the two-sided significance level of 0.0037 calculated by Lan-DeMets alpha spending function with an O'Brien-Fleming boundary.

Results of additional interim analysis of OS (ITT population, data cut-off date of July 31, 2012)

	T-DM1 group	Lapatinib/Cape group
Number of patients	495	496
Number of deaths (%)	149 (30.1)	182 (36.7)
Median OS [95% CI] (months)	30.9 [26.81, 34.27]	25.1 [22.74, 27.96]
Hazard ratio [95% CI][*1]	0.682 [0.548, 0.849]	
P value (two-sided)[*2]	0.0006	

*1: Based on the Cox proportional hazard model stratified by number of chemotherapy regimens (0-1 vs. >1), presence or absence of visceral metastasis, world region (US, Western Europe, other), *2: Log-rank test stratified by the number of chemotherapy regimens (0-1 vs. >1), presence or absence of visceral metastasis, world region (US, Western Europe, other), two-sided significance level 0.0037



Results of additional interim analysis of OS (ITT population, data cut-off date of July 31, 2012)

As regards safety, death within 30 days after the last dose of the study drug was reported in 4 patients in the T-DM1 group and in 17 patients in the lapatinib/Cape group. The deaths were caused by disease progression in 3 patients in the T-DM1 group and in 13 patients in the lapatinib/Cape group, whereas the causes of death in other patients were metabolic encephalopathy in 1 patient in the T-DM1 group and coronary artery disease, hydrocephalus, coma, and multi-organ failure in 1 patient each in the lapatinib/Cape group. Among these fatal adverse events, a causal relationship to the study drug could not be ruled out for metabolic encephalopathy in 1 patient in the T-DM1 group and coronary artery disease and multi-organ failure in 1 patient each in the lapatinib/Cape group.

Reference data

(1) Foreign clinical studies

1) Foreign phase I/II study (5.3.5.4.1, Study TDM4373g/Study BO22495 [[redacted] to [redacted]])

An open-label, uncontrolled study was conducted to evaluate the safety and tolerability of concomitant use of T-DM1 with pertuzumab (T-DM1/pertuzumab group) in patients with HER2-positive metastatic or recurrent breast cancer who had not previously been treated with chemotherapy or who progressed after chemotherapy including HER2-targeted treatment (target sample size, 60) in 17 medical institutions overseas.

Of 67 patients enrolled in the study, 64 patients received the study drug and were included in the safety analysis. Death was reported in 1 patient within 30 days after the last dose of the study drug. The death was caused by pneumonia and its causal relationship to the study drug was ruled out.

2) Foreign phase II study (5.3.5.1.2, Study TDM4450g/Study BO21976 [September 2008 to August 2011])

An open-label, randomized, comparative study was conducted to evaluate the efficacy and safety of T-DM1 in patients with HER2-positive metastatic or recurrent breast cancer who had not previously been treated with chemotherapy except neoadjuvant or adjuvant chemotherapy (target sample size, 120) in 65 medical institutions overseas. The trastuzumab/DTX group was included as the control group.

Of 137 patients enrolled in the study (67 patients in the T-DM1 group, 70 patients in the trastuzumab/DTX group), 135 patients who received the study drug (66 patients in the T-DM1 group, 69 patients in the trastuzumab/DTX group) were included in the safety analysis. Death within 30 days after the last dose of the study drug was reported in 2 patients (2.9%) in the T-DM1 group and in 1 patient (1.5%) in the trastuzumab/DTX group. One patient in the T-DM1 group died of disease progression, whereas the causes of deaths in other patients were sudden death in the T-DM1 group and cardiopulmonary failure in the trastuzumab/DTX group. A causal relationship to the study drug was ruled out for both events.

3) Foreign phase II study (5.3.4.2.1, Study TDM4688g [[redacted] to [redacted]])

An open-label, uncontrolled study was conducted to evaluate the effect of T-DM1 on corrected QT interval in patients with HER2-positive metastatic or recurrent breast cancer who progressed after chemotherapy including trastuzumab (target sample size, 50), and to evaluate the safety and tolerability of concomitant use of T-DM1 with pertuzumab in patients who progressed during the early stage of T-DM1 monotherapy in 13 medical institutions overseas.

All of the 51 patients enrolled in the study received T-DM1. There were no deaths up to 30 days after the last dose of the study drugs.

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

4.(iii).B.(1) Data for review

PMDA concluded that, among the submitted evaluation data, the most important clinical study for evaluating the efficacy and safety of T-DM1 was the foreign phase III study (EMILIA study) which evaluated the efficacy and safety of T-DM1 in patients with HER2-positive metastatic or recurrent breast cancer who had previously been treated with chemotherapy with a taxane antineoplastic drug and trastuzumab. Thus, PMDA decided to evaluate the submitted data focused on the EMILIA study.

As regards the efficacy and safety of T-DM1 in Japanese patients, PMDA decided to evaluate the submitted data focusing on the Japanese phase II study (Study JO22997) which was conducted in Japanese patients with HER2-positive metastatic or recurrent breast cancer using the same dosage regimen as in the EMILIA study.

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

Based on the following review, PMDA has concluded that T-DM1 is effective in patients with HER2-positive, inoperable or recurrent breast cancer.

4.(iii).B.(2).1) Use of control group

PMDA asked the applicant to explain the reason for using lapatinib/Cape group as the control in the EMILIA study.

The applicant responded as follows:

When the EMILIA study started in February 2009, the US National Comprehensive Cancer Network Clinical Practice Guidelines in Oncology, Breast Cancer (NCCN guidelines) (v.1.2009) stated that the first-line therapy for patients with HER2-positive inoperable or recurrent breast cancer was concomitant use of trastuzumab with other antineoplastic drugs. Also, one of the standard treatments recommended by the guideline in the case of disease progression during or after trastuzumab administration was either the combination of lapatinib and capecitabine or combination of trastuzumab with an antineoplastic drug that was not used concomitantly in the primary treatment.

In patients who have received the above trastuzumab therapy as neoadjuvant or adjuvant chemotherapy, re-administration of trastuzumab is recommended only if recurrence occurres at least 6 months after the therapy (*Curr Oncol*. 2009;16:25-35). Thus, lapatinib/Cape combination therapy was considered to be the priority treatment for patients who had a relapse within a short period after trastuzumab therapy.

On the basis of the above, the lapatinib/Cape group was used as the control group in the EMILIA study in patients who had previously been treated with chemotherapy with a taxane antineoplastic drug and trastuzumab.

PMDA accepted the applicant's response.

4.(iii).B.(2).2) Efficacy endpoints and results of efficacy evaluation

PMDA considers as follows:

It is appropriate to use OS as one of the primary efficacy endpoints in the EMILIA study in patients with HER2-positive metastatic or recurrent breast cancer who had previously been treated with chemotherapy with a taxane antineoplastic drug and trastuzumab, because OS is the true endpoint in such patients.

In the EMILIA study, superiority of the T-DM1 group to the lapatinib/Cape group in PFS was confirmed, and a significant improvement in OS was shown as well, as determined by the results of the additional interim analysis (*P* value) [see "4.(iii).A. Evaluation data (2)4) Foreign phase III study"]. Thus, PMDA concluded that T-DM1 is effective in the target patients in this study. Also, a certain level of response rate was obtained in the Japanese phase II study (Study JO22997) in which T-DM1 was administered according to the same dosage regimen as in the EMILIA study [see "4.(iii).A. Evaluation data (1)2) Japanese phase II study"]. Therefore, PMDA considers that T-DM1 is effective in Japanese patients as well, by taking account of the data of the EMILIA study.

4.(iii).B.(3) Safety [for adverse events, see "4.(iv) Adverse events, etc. observed in clinical studies"]

As a result of the reviews described below, PMDA considers that caution is required in administering T-DM1 for the following adverse events: hepatotoxicity, nodular regenerative hyperplasia, thrombocytopenia, infusion reaction, interstitial lung disease, cardiac function failed, and neuropathy peripheral.

PMDA has concluded that T-DM1 is tolerable provided that appropriate measures such as monitoring and control of adverse events as well as the treatment withdrawal, dose reduction, or discontinuation of T-DM1 are taken by physicians with sufficient knowledge and experience of cancer chemotherapy.

4.(iii).B.(3).1) Safety profile and its differences between Japanese and foreign patients

The outlines of safety profile in the EMILIA study and Study JO22997 were as shown in the following table.

Summary of safety (EMILIA study)

	Number of patients (%)	
	T-DM1 group N = 490	Lapatinib/Cape group N = 488
All adverse events	470 (95.9)	477 (97.7)
Adverse events of Grade ≥3	200 (40.8)	278 (57.0)
Serious adverse events	76 (15.5)	88 (18.0)
Adverse events leading to treatment discontinuation	29 (5.9)	Lapatinib 37 (7.6) Cape 46 (9.4)
Adverse events leading to dose reduction	74 (15.1)	Lapatinib 92 (18.9) Cape 188 (38.5)
Adverse events leading to treatment interruption	104 (21.2)	Lapatinib 180 (36.9) Cape 214 (43.9)

Summary of safety (Study JO22997)

	Number of patients (%) N = 73
All adverse events	70 (95.9)
Adverse events of Grade ≥3	41 (56.2)
Serious adverse events	15 (20.5)
Adverse events leading to treatment discontinuation	14 (19.2)
Adverse events leading to dose reduction	22 (30.1)
Adverse events leading to treatment interruption	38 (52.1)

Based on the safety information obtained from the T-DM1 group and the lapatinib/Cape group in the EMILIA study, the applicant explained the safety profile of T-DM1 as follows:
Among adverse events with an incidence of ≥10% of patients in the T-DM1 group, the Grade ≥3 event with an incidence of ≥5% was thrombocytopenia (12.9%). Among adverse events with an incidence of ≥10% of patients in the lapatinib/Cape group, Grade ≥3 events with an incidence of ≥5% were diarrhoea (20.7%) and palmar-plantar erythrodysaesthesia syndrome (16.4%). Adverse events of Grade ≥3 that occurred with ≥2% higher incidence in the T-DM1 group compared with the lapatinib/Cape group were thrombocytopenia (12.9% in the T-DM1 group, 0.2% in the lapatinib/Cape group) and AST increased (4.3%, 0.8%). Adverse events of Grade ≥3 that occurred with ≥2% higher incidence in the lapatinib/Cape group compared with the T-DM1 group were diarrhoea (1.6%, 20.7%), palmar-plantar erythrodysaesthesia syndrome (0%, 16.4%), vomiting (0.8%, 4.5%), mucosal inflammation (0.2%, 2.3%), and neutropenia (2.0%, 4.3%) (see table below).

Adverse events with an incidence of ≥10% in either group (EMILIA study)

Event	Number of patients (%)			
	T-DM1 group N = 490		Lapatinib/Cape group N = 488	
	All Grades	Grade ≥3	All Grades	Grade ≥3
All adverse events	470 (95.9)	200 (40.8)	477 (97.7)	278 (57.0)
Nausea	192 (39.2)	4 (0.8)	218 (44.7)	12 (2.5)
Fatigue	172 (35.1)	12 (2.4)	136 (27.9)	17 (3.5)
Thrombocytopenia	137 (28.0)	63 (12.9)	12 (2.5)	1 (0.2)
Headache	133 (27.1)	4 (0.8)	68 (13.9)	4 (0.8)
Constipation	124 (25.3)	2 (0.4)	47 (9.6)	0
Diarrhoea	114 (23.3)	8 (1.6)	389 (79.7)	101 (20.7)
AST increased	110 (22.4)	21 (4.3)	46 (9.4)	4 (0.8)
Decreased appetite	101 (20.6)	2 (0.4)	113 (23.2)	5 (1.0)
Epistaxis	99 (20.2)	1 (0.2)	39 (8.0)	0
Vomiting	93 (19.0)	4 (0.8)	143 (29.3)	22 (4.5)
Asthenia	86 (17.6)	2 (0.4)	81 (16.6)	7 (1.4)
Arthralgia	85 (17.3)	3 (0.6)	38 (7.8)	0
Pyrexia	85 (17.3)	0	37 (7.6)	2 (0.4)
ALT increased	83 (16.9)	14 (2.9)	43 (8.8)	7 (1.4)
Cough	83 (16.9)	1 (0.2)	60 (12.3)	1 (0.2)
Dry mouth	77 (15.7)	0	24 (4.9)	1 (0.2)
Myalgia	69 (14.1)	3 (0.6)	18 (3.7)	0
Back pain	64 (13.1)	3 (0.6)	50 (10.2)	2 (0.4)
Abdominal pain upper	57 (11.6)	2 (0.4)	41 (8.4)	1 (0.2)
Dyspnoea	56 (11.4)	3 (0.6)	36 (7.4)	2 (0.4)
Insomnia	54 (11.0)	2 (0.4)	41 (8.4)	1 (0.2)
Pain in extremity	52 (10.6)	2 (0.4)	52 (10.7)	5 (1.0)
Rash	52 (10.6)	0	130 (26.6)	9 (1.8)
Anaemia	51 (10.4)	13 (2.7)	39 (8.0)	8 (1.6)
Neuropathy peripheral	49 (10.0)	8 (1.6)	28 (5.7)	1 (0.2)
Dizziness	48 (9.8)	1 (0.2)	51 (10.5)	1 (0.2)
Dyspepsia	43 (8.8)	0	56 (11.5)	2 (0.4)
Mucosal inflammation	33 (6.7)	1 (0.2)	93 (19.1)	11 (2.3)
Dry skin	17 (3.5)	0	49 (10.0)	1 (0.2)
Stomatitis	16 (3.3)	0	61 (12.5)	2 (0.4)
Palmar-plantar erythrodysaesthesia syndrome	6 (1.2)	0	283 (58.0)	80 (16.4)
Paronychia	1 (0.2)	0	52 (10.7)	3 (0.6)

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase

PMDA asked the applicant to explain the differences in safety of T-DM1 between Japanese and foreign patients.

The applicant responded as follows:
In the T-DM1 group of the EMILIA study and in Study JO22997, adverse events with an incidence of ≥10% and Grade ≥3 adverse events were as shown in the following table. Adverse events that occurred with ≥10% higher incidence in Study JO22997 than in the T-DM1 group of the EMILIA study were platelet count decreased, neutrophil count decreased, malaise, stomatitis, epistaxis, pyrexia, and nasopharyngitis. Adverse events that occurred with ≥10% higher incidence in the T-DM1 group of the EMILIA study than in Study JO22997 were thrombocytopenia, fatigue, diarrhoea, asthenia, and dyspnoea. The evaluation of combined data of adverse events related to decrease in platelet count (thrombocytopenia, platelet count decreased, platelet disorder) showed that there were no significant differences in the incidence between Study JO22997 and the T-DM1 group of the EMILIA study.

Among adverse events with an incidence of ≥10%, Grade ≥3 adverse events that occurred with ≥5% higher incidence in Study JO22997 than in the T-DM1 group of the EMILIA study were thrombocytopenia*, AST increased, and ALT increased. The incidence of serious adverse events

was 15.5% in the T-DM1 group of the EMILIA study and 20.5% in Study JO22997. In Study JO22997, serious adverse events occurred in 1 patient each (1.4%) except malaise which occurred in 2 patients (2.7%). There were no serious adverse events with significantly different incidence between the T-DM1 group of the EMILIA study and Study JO22997.

*: Adverse events related to decrease in platelet count (thrombocytopenia, platelet count decreased, platelet disorder) were combined together and defined as thrombocytopenia.

Adverse events with an incidence of ≥10% in either group

Event	Number of patients (%)			
	EMILIA study T-DM1 group N = 490		Study JO22997 N = 73	
	All Grades	Grade ≥3	All Grades	Grade ≥3
All adverse events	470 (95.9)	200 (40.8)	70 (95.9)	41 (56.2)
Nausea	192 (39.2)	4 (0.8)	32 (43.8)	0
Fatigue	172 (35.1)	12 (2.4)	10 (13.7)	1 (1.4)
Thrombocytopenia	137 (28.0)	63 (12.9)	0	0
Headache	133 (27.1)	4 (0.8)	14 (19.2)	0
Constipation	124 (25.3)	2 (0.4)	12 (16.4)	0
Diarrhoea	114 (23.3)	8 (1.6)	6 (8.2)	0
AST increased	110 (22.4)	21 (4.3)	15 (20.5)	10 (13.7)
Decreased appetite	101 (20.6)	2 (0.4)	22 (30.1)	2 (2.7)
Epistaxis	99 (20.2)	1 (0.2)	30 (41.1)	0
Vomiting	93 (19.0)	4 (0.8)	14 (19.2)	4 (5.5)
Asthenia	86 (17.6)	2 (0.4)	0	0
Arthralgia	85 (17.3)	3 (0.6)	6 (8.2)	0
Pyrexia	85 (17.3)	0	26 (35.6)	0
ALT increased	83 (16.9)	14 (2.9)	8 (11.0)	6 (8.2)
Cough	83 (16.9)	1 (0.2)	8 (11.0)	0
Dry mouth	77 (15.7)	0	6 (8.2)	0
Myalgia	69 (14.1)	3 (0.6)	3 (4.1)	0
Back pain	64 (13.1)	3 (0.6)	6 (8.2)	0
Abdominal pain upper	57 (11.6)	2 (0.4)	5 (6.8)	0
Dyspnoea	56 (11.4)	3 (0.6)	0	0
Insomnia	54 (11.0)	2 (0.4)	3 (4.1)	0
Pain in extremity	52 (10.6)	2 (0.4)	2 (2.7)	0
Rash	52 (10.6)	0	10 (13.7)	0
Anaemia	51 (10.4)	13 (2.7)	3 (4.1)	2 (2.7)
Neuropathy peripheral	49 (10.0)	8 (1.6)	4 (5.5)	0
Nasopharyngitis	41 (8.4)	0	24 (32.9)	0
Malaise	3 (0.6)	0	23 (31.5)	0
Platelet count decreased	14 (2.9)	5 (1.0)	20 (27.4)	16 (21.9)
Stomatitis	16 (3.3)	0	12 (16.4)	0
Neutrophil count decreased	4 (0.8)	0	10 (13.7)	2 (2.7)
Peripheral sensory neuropathy	29 (5.9)	5 (1.0)	8 (11.0)	1 (1.4)
Chills	39 (8.0)	0	8 (11.0)	0

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase

PMDA considers as follows:
In the EMILIA study, each incidence of all the adverse events, Grade ≥3 adverse events, serious adverse events, and adverse events leading to treatment discontinuation tended to be lower in the T-DM1 group than in the lapatinib/Cape group. Thus, it is concluded that T-DM1 is tolerable.

However, caution should be exercised against the events that occurred with a higher incidence in the T-DM1 group than in the lapatinib/Cape group. Among such events, particular attention should be paid to hepatotoxicity and thrombocytopenia because those events of Grade ≥3 occurred more in the T-DM1 group than in the lapatinib/Cape group. Although there is a limitation to the

comparison of the safety profile of T-DM1 between the Japanese patients in Study JO22997 and the foreign patients in the EMILIA study, particular attention should be paid, in using T-DM1 in Japan, to the events that occurred with a higher incidence in Japanese patients in clinical studies. Information on the incidence of these events should be appropriately provided to clinical practice. Furthermore, because of the limited number of Japanese patients who have been treated with T-DM1, safety information on T-DM1 in Japanese patients has not been sufficiently accumulated. Therefore, safety information in Japanese patients should be collected after the market launch [see "4.(iii).B.(6) Post-marketing investigations"].

4.(iii).B.(3).2) Hepatotoxicity and nodular regenerative hyperplasia

The applicant explained hepatotoxicity associated with T-DM1 as follows:

In the EMILIA study, the incidence of hepatotoxicity (events corresponding to "cholestasis and jaundice of hepatic origin," "liver related investigations, signs and symptoms," "drug related hepatic disorders - severe events only [hepatic failure, fibrosis and cirrhosis and other liver damage-related conditions," "hepatitis, non-infectious," "liver neoplasms, benign [incl cysts and polyps]," "liver neoplasms, malignant and unspecified"] in MedDRA Standardised MedDRA Queries [SMQ]) was 31.0% (152 of 490 patients) in the T-DM1 group and 25.2% (123 of 488 patients) in the lapatinib/Cape group. Of these, the incidence of Grade ≥3 events was 8.8% (43 of 490 patients) in the T-DM1 group and 4.7% (23 of 488 patients) in the lapatinib/Cape group. The incidence of hepatotoxicity leading to treatment discontinuation was 1.8% (9 of 490 patients) in the T-DM1 group and 0% (0 of 488 patients) in the lapatinib/Cape group. The incidence of serious adverse events was 0.8% (4 of 490 patients) in the T-DM1 group and 0.2% (1 of 488 patients) in the lapatinib/Cape group.

The incidence of hepatotoxicity in all the patients* treated with T-DM1 in clinical studies on T-DM1 was 31.9% (305 of 955 patients) and the incidence in the Japanese patients in Study JO22997 was 34.2% (25 of 73 patients). The incidence of Grade ≥3 hepatotoxicity was 9.6% (92 of 955 patients) in all the patients and 20.5% (15 of 73 patients) in the Japanese patients. The incidence of hepatotoxicity leading to treatment discontinuation was 2.1% (20 of 955 patients) in all the patients and 5.5% (4 of 73 patients) in the Japanese patients. The incidence of serious adverse events was 1.0% (10 of 955 patients) in all the patients and 1.4% (1 of 73 patients) in the Japanese patients, and 3 patients of them were diagnosed with nodular regenerative hyperplasia. Among patients with serious adverse events, 2 patients died because of hepatic function abnormal and hepatic failure, respectively. A causal relationship to the study drug could not be ruled out for both events.

*: Patients who received T-DM1 alone (3.6 mg/kg every 3 weeks) in Japanese or foreign clinical studies (EMILIA study, Studies TDM4258g, TDM4374g, TDM3569g, TDM4450g, TDM4688g, TDM4529g, and JO22997) The same applies hereinafter.

The applicant explained nodular regenerative hyperplasia associated with T-DM1 administration as follows:

The safety information was assessed based on the events reported in the safety database of F. Hoffmann-La Roche, Ltd. since the initiation of clinical studies in 2006 up to [redacted] (in foreign patients) and on the events reported until [redacted] (in Japanese patients). As a result, nodular regenerative hyperplasia was diagnosed by liver biopsy in 5 foreign patients and in 1 Japanese patient, for none of which a causal relationship with the study drug could be ruled out. Of these, 1 foreign patient died of hepatic failure (the table below). This patient was diagnosed as fulfilling Hy's law (ALT >3 times the upper limit of normal and total bilirubin ≥2 times the upper limit of normal, or ALT >8 times the upper limit of normal) after the above data cut-off date, but the patient was ruled out for severe acute drug-induced liver injury (DILI).

All of the 6 patients underwent liver biopsy and were diagnosed with nodular regenerative

hyperplasia based on the histopathological findings.

Details of patients diagnosed with nodular regenerative hyperplasia

Patient No.	Reported event name	Time of onset (months)	Portal hypertension symptoms	Treatment	Outcome
1	Hepatic cirrhosis	15	Splenomegaly, ascites, varices oesophageal	T-DM1 discontinued Portal hypertension treated	Improved
2	Hepatic failure	25	Ascites	None (died before the scheduled next administration)	Death
3	Blood bilirubin increased	2.5	None	T-DM1 discontinued	Unknown
4	Portal hypertension	16	Splenomegaly, varices oesophageal	T-DM1 discontinued Portal hypertension treated	Unknown
5	Portal hypertension	16	Gastrooesophageal varices	T-DM1 discontinued Gastrooesophageal varices treated	Unrecovered
6	Nodular regenerative hyperplasia	22	Splenomegaly	None (T-DM1 continued)	Unrecovered

Risk factor(s) for T-DM1-induced nodular regenerative hyperplasia are currently unknown. However, in light of the findings that nodular regenerative hyperplasia was diagnosed in 5 of 6 patients after ≥1 year of administration of T-DM1, the risk may be higher in patients who are treated with T-DM1 for a long period. The mechanism of the onset of nodular regenerative hyperplasia is generally unknown, and it is the same with T-DM1-induced nodular regenerative hyperplasia. However, in light of the findings that hepatotoxicity developed after T-DM1 administration in some patients, T-DM1 may possibly induce nodular regenerative hyperplasia.

PMDA considers as follows:
In the T-DM1 group in the EMILIA study, the incidence of hepatotoxicity, including Grade ≥3 events, tended to be higher than in the lapatinib/Cape group, and the incidence of hepatotoxicity leading to treatment discontinuation also tended to be slightly higher. In Japanese patients, the incidences of Grade ≥3 events and of hepatotoxicity leading to treatment discontinuation tended to be higher compared with all the patients receiving T-DM1 in the clinical studies on T-DM1. In addition, there were patients who died of serious hepatotoxicity (hepatic function abnormal, hepatic failure) in foreign clinical studies. Therefore, it is necessary to caution that patients should be monitored through liver function tests and, if any abnormalities are observed, appropriate measures such as treatment interruption, dose reduction, or treatment discontinuation should be taken [see “4.(iii).B.(5).2) Criteria for treatment interruption, dose reduction, and treatment discontinuation”].

After T-DM1 administration, nodular regenerative hyperplasia occurred and resulted in death in some patients. In addition, patients with the disease may present with symptoms of portal hypertension that can become aggravated. Therefore, caution should be exercised against the possible occurrence of nodular regenerative hyperplasia during T-DM1 administration. In some of the above 6 patients, no significant changes were observed in the laboratory values related to liver (e.g., ALT, bilirubin); instead, portal hypertension symptoms such as splenomegaly and varicose vein were signs that led to the diagnosis of nodular regenerative hyperplasia. Generally, the disease can be determined only by liver biopsy. Therefore, it is necessary to caution that patients should be monitored for portal hypertension symptoms and imaging findings during T-DM1 administration and, if portal hypertension or uneven hepatic parenchymal findings are suspected, liver biopsy should be considered.

4.(iii).B.(3).3) Thrombocytopenia and haemorrhage

The applicant explained T-DM1-induced thrombocytopenia and haemorrhage as follows:

In the EMILIA study, the incidence of thrombocytopenia (events corresponding to "thrombocytopenia" in MedDRA SMQ) was 30.4% (149 of 490 patients) in the T-DM1 group and 2.9% (14 of 488 patients) in the lapatinib/Cape group. The incidence of Grade ≥3 events was 13.9% (68 of 490 patients) in the T-DM1 group and 0.2% (1 of 488 patients) in the lapatinib/Cape group. The incidence of thrombocytopenia leading to treatment discontinuation was 2.0% (10 of 490 patients) in the T-DM1 group and 0% (0 of 488 patients) in the lapatinib/Cape group, and the incidence of serious adverse events was 0.6% (3 of 490 patients) in the T-DM1 group and 0.2% (1 of 488 patients) in the lapatinib/Cape group.

The incidence of thrombocytopenia in all the patients receiving T-DM1 in the clinical studies on T-DM1 was 31.2% (298 of 955 patients) and the incidence in Japanese patients in Study JO22997 was 27.4% (20 of 73 patients). The incidence of Grade ≥3 thrombocytopenia was 12.1% (116 of 955 patients) in all the patients and 21.9% (16 of 73 patients) in the Japanese patients. The incidence of thrombocytopenia leading to treatment discontinuation was 1.6% (15 of 955 patients) in all the patients and 1.4% (1 of 73 patients) in the Japanese patients, and the incidence of serious adverse events was 0.7% (7 of 955 patients) in all the patients and 0% (0 of 73 patients) in the Japanese patients.

In the EMILIA study, the incidence of haemorrhage (events corresponding to "haemorrhage laboratory terms [narrow]" and "haemorrhage terms [excl laboratory terms]" in MedDRA SMQ) was 29.8% (146 of 490 patients) in the T-DM1 group and 15.8% (77 of 488 patients) in the lapatinib/Cape group. The incidence of Grade ≥3 events was 1.4% (7 of 490 patients) in the T-DM1 group and 0.8% (4 of 488 patients) in the lapatinib/Cape group. There was no patient who discontinued the administration because of haemorrhage. The incidence of serious adverse events was 1.2% (6 of 490 patients) in the T-DM1 group and 0.8% (4 of 488 patients) in the lapatinib/Cape group.

The incidence of haemorrhage in all the patients receiving T-DM1 in the clinical studies on T-DM1 was 35.6% (340 of 955 patients) and the incidence in the Japanese patients in Study JO22997 was 53.4% (39 of 73 patients). The incidence of Grade ≥3 haemorrphage was 1.7% (16 of 955 patients) in all the patients and 1.4% (1 of 73 patients) in the Japanese patients. The main event was Grade 1 or 2 epistaxis. There was no patient who discontinued administration because of haemorrhage. The incidence of serious adverse events was 1.3% (12 of 955 patients) in all the patients and 1.4% (1 of 73 patients) in the Japanese patients.

PMDA considers that caution should be provided to take appropriate measures such as dose reduction and treatment interruption against thrombocytopenia [see "4.(iii).B.(5).2) Criteria for treatment interruption, dose reduction, and treatment discontinuation"], for the following reasons.

- In the EMILIA study, the incidence of thrombocytopenia, including that of Grade ≥3 events, tended to be higher in the T-DM1 group than in the lapatinib/Cape group.
- The incidence of Grade ≥3 events was higher in the Japanese patients compared with all the patients receiving T-DM1 in the clinical studies on T-DM1.
- The relationship between thrombocytopenia and haemorrhage caused by T-DM1 is unclear. However, severe thrombocytopenia generally has a risk of haemorrhage. In addition, the incidence of haemorrhage in the EMILIA study, including that of Grade ≥3 events, tended to be higher in the T-DM1 group than in the lapatinib/Cape group.

4.(iii).B.(3).4) Infusion reaction

The applicant explained T-DM1-induced infusion reaction as follows:

In the EMILIA study, the incidence of infusion-related reaction/hypersensitivity (events corresponding to "anaphylactic reaction [narrow]" and "angioedema [narrow]" in MedDRA SMQ and "infusion related reaction" and "hypersensitivity" in MedDRA Preferred Terms) was 3.9% (19 of 490 patients). No Grade ≥3 events were observed. The incidence of infusion-related reaction/hypersensitivity leading to treatment discontinuation was 0.2% (1 of 490 patients), and the incidence of serious adverse events was 0.4% (2 of 490 patients). Infusion-related reaction/hypersensitivity observed in 19 patients in the EMILIA study included infusion-related reaction occurred in 7 patients (1.4%), hypersensitivity in 4 patients (0.8%), and pharyngeal oedema in 1 patient (0.2%) in Cycle 1. All of them occurred on the day of the first dose or the next day. In subsequent cycles, infusion-related reaction/hypersensitivity occurred in 1 patient in Cycle 2, 2 patients in Cycle 3, 1 patient each in Cycles 6 to 9, and 2 patients in Cycle 10 (either of face oedema, urticaria, infusion related reaction, or hypersensitivity, including patients with multiple events).

The incidence of infusion-related reaction/hypersensitivity in all the patients receiving T-DM1 in the clinical studies on T-DM1 was 6.5% (62 of 955 patients) and the incidence in Japanese patients in Study JO22997 was 4.1% (3 of 73 patients). The incidence of Grade ≥3 events was 0.1% (1 of 955 patients) in all the patients and 0% in the Japanese patients. The incidence of infusion-related reaction/hypersensitivity leading to treatment discontinuation was 0.1% (1 of 955 patients) in all the patients and 0% in the Japanese patients. The incidence of serious adverse events was 0.2% (2 of 955 patients) in all the patients and 0% in the Japanese patients. The breakdown of infusion-related reaction/hypersensitivity observed in the 3 patients in Study JO22997 was urticaria in 1 patient (1.4%) in Cycle 1, urticaria and face oedema in 1 patient each (1.4%) in Cycle 2, and lip oedema in 1 patient (1.4%) in Cycle 16 (including patients with multiple events).

PMDA asked the applicant to explain premedication to prevent T-DM1-induced infusion reaction.

The applicant responded as follows:

In the EMILIA study, use of steroid as a premedication before the initial dose of T-DM1 was allowed if the medical monitor of the sponsor approved. Premedication to prevent nausea and anxiety was allowed as well. In the EMILIA study in which T-DM1 was administered to 490 patients, infusion reaction was observed in 4 of 87 patients who used approved combination drugs for prophylactic use on the first day of T-DM1 dosing and in 8 of 403 patients who did not receive premedication. When reactions reported in Cycle 2 and subsequent cycles were included, the number of patients who developed infusion reaction was 19. Nine of these patients were given prophylactic premedication in the next cycle of the reaction. Of these 19 patients, 1 patient who experienced infusion-related reaction had the same event twice after the first infusion reaction, whereas in the remaining 18 patients, no other infusion reactions were reported in the subsequent cycles regardless of use or non-use of premedication.

In Study JO22997, prophylactic use of premedication was prohibited against yet-to-be observed adverse events, including infusion reaction. Therefore, prophylactic use of premedication against infusion reaction was not given to patients who had not experienced the event. In the study, urticaria, face oedema, and lip oedema, which were adverse events defined as infusion reactions, occurred in 1 patient each, but the patients with the adverse events had received no prophylactic premedication before the inital onset of infusion reaction. Of these, 1 patient who had experienced urticarial was given prophylactic premedication in the next cycle, but developed urticaria in this cycle as well. In the remaining 2 patients, no premedication was given in any of the cycles, but no further infusion reaction was observed.

Next, PMDA asked the applicant to explain the measures against infusion reaction following T-DM1 administration.

The applicant responded as follows:
In the EMILIA study, if T-DM1-related increased body temperature of >38.5°C or other mild infusion-associated symptoms occurred after the occurrence of infusion reaction, (a) patients could be treated with acetaminophen or H_1 or H_2 receptor antagonists (e.g., diphenhydramine hydrochloride [diphenhydramine], ranitidine), depending on the symptom, and (b) serious infusion-related events associated with dyspnoea, hypotension, wheezing, bronchospasm, tachycardia, hypoxia, or respiratory distress were to be managed by symptomatic treatment (e.g., oxygen inhalation, β-agonist, corticosteroid) according to the standard clinical procedure. In the EMILIA study, infusion reaction occurred in 19 patients and the adverse event was treated in 16 of the 19 patients. Of 21 infusion reactions that occurred in the 19 patients, 8 events resulted in treatment interruption of T-DM1, 3 events required adjustment of the speed of T-DM1 infusion, 1 event resulted in discontinuation of T-DM1, while no measures, including dose reduction, were taken for the remaining 9 events.

Study JO22997 had specified as follows regarding the measures against infusion reaction. There were 4 infusion reactions in 3 patients, but none of these patients were treated for the adverse event, or resulted in temporary withdrawal, dose reduction, or discontinuation of T-DM1.

Rules specified in Study JO22997

> If dyspnea or clinically marked hypotension occurs after the occurrence of infusion reaction, T-DM1 shall be temporarily withdrawn. If Grade 3 or 4 allergic reaction/hypersensitivity, adult respiratory distress syndrome, or bronchospasm (regardless of Grade) occurs, the patient should be treated with oxygen inhalation, β2 agonist, or corticosteroid as necessary, and further T-DM1 treatment should be discontinued. If other mild to moderate findings, such as pyrexia of ≥38.0°C, are observed, T-DM1 treatment can be continued at the infusion speed of ≤50% decrease or the treatment should be temporarily withdrawn. The patient should be treated with acetaminophen, diphenhydramine, or ranitidine and monitored until the clinically significant findings have completely resolved. If there is no problem in tolerability, the infusion speed may be increased by 50% in every 30 minutes. In subsequent cycles, the infusion may be resumed at a 100% speed with due caution. Acetaminophen or diphenhydramine may be given before the administration of the study drug.

PMDA considers as follows:
Infusion reactions were observed after treatment of T-DM1, both in the Japanese and foreign clinical studies. In Study JO22997, T-DM1 treatment could be continued without any particular measures being taken. In the EMILIA study, infusion reactions were tolerable by measures such as treatment interruption, reduction in infusion speed, and treatment discontinuation. Taking account of these situations, it should be cautioned that if infusion reaction is observed, appropriate measures such as discontinuation of T-DM1 should be taken. Also, patient conditions should be monitored by taking notice of the fact that the first infusion reaction may occur in Cycle 2 or subsequent cycles. The significance of premedication against infusion reaction is unclear from the results of the clinical studies currently available. Thus, there is no justification for recommending premedication.

4.(iii).B.(3).5) Interstitial lung disease

The applicant explained T-DM1-induced interstitial lung disease as follows:
In the EMILIA study, the incidence of events corresponding to "Interstitial lung disease (narrow)" in MedDRA SMQ was 1.2% (6 of 490 patients, pneumonitis in each patient) in the T-DM1 group

and 0.4% (2 of 488 patients, alveolitis allergic and radiation pneumonitis in 1 patient each) in the lapatinib/Cape group. No Grade ≥3 events were observed in either of the groups. Treatment was discontinued only in 1 of 490 patients in the T-DM1 group (0.2%, pneumonitis). The incidence of serious adverse events was 0.2% (1 of 490 patients, pneumonitis) in the T-DM1 group and 0.2% (1 of 488 patients, allergic alveolitis) in the lapatinib/Cape group.

The incidence of events in all the patients receiving T-DM1 in the clinical studies on T-DM1 was 0.9% (9 of 955 patients, pneumonitis in 8 patients, interstitial lung disease in 2 patients [including 1 patient with multiple adverse events]). The events in 4 patients were assessed as not causally related to the study drug. The incidence of Grade ≥3 events was 0.2% (2 of 955 patients, pneumonitis and interstitial lung disease in 1 patient each). Pneumonitis in 1 patient was Grade 3 (assessed as causally related), and interstitial lung disease in 1 patient was Grade 5 (death, assessed as not causally related). In the fatal case, the patient received T-DM1 (3.6 mg/kg) through Day 278, was diagnosed with interstitial lung disease on Day 309, and died on Day 314 because of respiratory failure caused by interstitial lung disease. T-DM1 treatment was discontinued in 1 of 955 patients (0.1%, pneumonitis), and the incidence of serious adverse events was 0.3% (3 of 955 patients, pneumonitis in 2 patients, interstitial lung disease in 1 patient). No interstitial lung disease was observed in Study JO22997 conducted in Japanese patients.

PMDA considers as follows:
In the EMILIA study, no significant differences were observed in the incidence of interstitial lung disease between the T-DM1 group and the lapatinib/Cape group. The disease was not observed in any of the Japanese patients. However, since 1 patient died because of the interstitial lung disease, caution should be exercised against the disease caused by T-DM1.

4.(iii).B.(3).6) Cardiac function failed

The applicant explained T-DM1-induced cardiac function failed as follows:
In the EMILIA study, the incidence of cardiac function failed (events corresponding to "cardiac failure [narrow]" in MedDRA SMQ) was 0.8% (4 of 490 patients, ejection fraction decreased in all the patients) in the T-DM1 group and 2.3% (11 of 488 patients, ejection fraction decreased in 10 patients, pulmonary oedema in 1 patient) in the lapatinib/Cape group. The incidence of Grade ≥3 events was 0% (0 of 490 patients) in the T-DM1 group and 0.6% (3 of 488 patients) in the lapatinib/Cape group. The incidence of cardiac function failed leading to treatment discontinuation was 0% (0 of 490 patients) in the T-DM1 group and 0.2% (1 of 488 patients) in the lapatinib/Cape group. The incidence of serious adverse events was 0% (0 of 490 patients) in the T-DM1 group and 0.2% (1 of 488 patients) in the lapatinib/Cape group.

The incidence of cardiac function failed in all the patients receiving T-DM1 in the clinical studies on T-DM1 was 1.5% (14 of 955 patients, ejection fraction decreased and pulmonary oedema), and the incidence in Japanese patients in Study JO22997 was 1.4% (1 of 73 patients, ejection fraction decreased). The incidence of Grade ≥3 events was 0.2% (2 of 955 patients) in all the patients and 0% (0 of 73 patients) in the Japanese patients. The incidence of cardiac function failed leading to treatment discontinuation was 0.2% (2 of 955 patients) in all the patients and 1.4% (1 of 73 patients) in the Japanese patients. No serious adverse events occurred in either of the groups.

By taking account of the occurrence of trastuzumab-induced cardiac function failed, left ventricular ejection fraction (LVEF) of ≥50% was listed in the inclusion criteria with regard to cardiac function in all Japanese and foreign clinical studies subjected to safety evaluation. Therefore, T-DM1 has never been administered to patients with LVEF of <50%. Also, periodical cardiac screening was required during the treatment with T-DM1 in all the studies. In the EMILIA study, cardiac screening was to be performed 6 and 12 weeks after the first dose, and every 12

weeks thereafter. In this study, cardiac function failed was observed in 4 patients. The failure occurred 29, 79, 155, and 169 days after the first dose, showing no consistent tendency regarding the time of occurrence. All of these events were Grade 1 or 2; none of them were serious or led to treatment discontinuation.

PMDA considers as follows:
In the EMILIA study, the incidence of cardiac function failed in the T-DM1 group was not higher than that in the lapatinib/Cape group. However, given that there is no experience of T-DM1 treatment to patients with cardiac function failed, and that cardiac function failed requires particular caution in using trastuzumab, it is necessary to raise caution against T-DM1-induced cardiac function failed. Also, by taking account of the periodical cardiac screening specified in the EMILIA study, Study JO22997, etc., it is appropriate to carry out cardiac screening before and during treatment with T-DM1 and to specify the criteria for treatment interruption and treatment discontinuation based on the decrease in LVEF [see "4.(iii).B.(5).2) Criteria for treatment interruption, dose reduction, and treatment discontinuation"].

4.(iii).B.(3).7) Neuropathy peripheral
The applicant explained T-DM1-induced neuropathy peripheral as follows:
In the EMILIA study, the incidence of neuropathy peripheral (events corresponding to "peripheral neuropathy" in MedDRA SMQ) was 23.3% (114 of 490 patients) in the T-DM1 group and 18.2% (89 of 488 patients) in the lapatinib/Cape group. The incidence of Grade ≥3 events was 3.1% (15 of 490 patients) in the T-DM1 group and 0.4% (2 of 488 patients) in the lapatinib/Cape group. The incidence of neuropathy peripheral leading to treatment discontinuation was 0.2% (1 of 490 patients) in the T-DM1 group and 0.2% (1 of 488 patients) in the lapatinib/Cape group. The incidence of serious adverse events was 0.2% (1 of 490 patients) in the T-DM1 group and 0% (0 of 488 patients) in the lapatinib/Cape group.

The incidence of neuropathy peripheral in all the patients receiving T-DM1 in the clinical studies on T-DM1 was 26.5% (253 of 955 patients), and the incidence in Japanese patients in Study JO22997 was 16.4% (12 of 73 patients). The incidence of Grade ≥3 events was 2.5% (24 of 955 patients) in all the patients and 1.4% (1 of 73 patients) in the Japanese patients. The incidence of neuropathy peripheral leading to treatment discontinuation was 0.4% (4 of 955 patients) in all the patients and 1.4% (1 of 73 patients) in the Japanese patients. The incidence of serious adverse events was 0.3% (3 of 955 patients) in all the patients and 1.4% (1 of 73 patients) in the Japanese patients.

In patients with neuropathy peripheral among all the patients receiving T-DM1 in the clinical studies on T-DM1, the median time (range) to the first occurrence of neuropathy peripheral was 65.0 days (1.0-642.0 days). For patients who have not recovered, the duration of persistence of neuropathy peripheral was 50.0 days (1.0-691.0 days) when the recovery date was assumed as the day of the last observation (limited to patients with data of the last observation day available) and 70.5 days (1.0-818.0 days) when the recovery date was assumed as the day of the last dose (in patients with data of the last observation day available, the recovery date was assumed to be the last observation day). At the cut-off date in each clinical study, neuropathy peripheral remained unresolved in 51.4% of patients.

PMDA considers as follows:
The incidence of neuropathy peripheral is higher in the T-DM1 group than in the lapatinib/Cape group, and it may take a long time to recover when it occures. Therefore, it should be cautioned that appropriate measures such as treatment interruption should be taken if neuropathy peripheral occurs [see "4.(iii).B.(5).2) Criteria for treatment interruption, dose reduction, and treatment discontinuation"].

4.(iii).B.(4) Clinical positioning and indications

The proposed indication for T-DM1 was "HER2-positive, inoperable or recurrent breast cancer." Also, the applicant explained at submission that the Precautions for Indications section of the package insert would include the following statement: (a) HER2 positivity testing should be conducted by a pathologist or laboratory with sufficient experience, (b) the efficacy and safety of T-DM1 in neoadjuvant or adjuvant chemotherapy have not been established, and (c) T-DM1 should be administered to patients who have received prior treatment with trastuzumab and a taxane antineoplastic drug.

Based on the results of reviews in "4.(iii).B.(2) Efficacy," "4.(iii).B.(3) Safety," and the following review, PMDA has concluded that the description of indication should be modified as "HER2-positive inoperable or recurrent breast cancer" and also the following caution statements should be included in the Precautions for Indications section of the package insert.

- T-DM1 should be administered to patients who have received prior treatment with trastuzumab and a taxane antineoplastic drug.

- HER2 positivity testing should be conducted by a pathologist or laboratory with sufficient experience.

- The efficacy and safety of T-DM1 in neoadjuvant or adjuvant chemotherapy have not been established.

4.(iii).B.(4).1) Clinical positioning of T-DM1 and patients to be treated

PMDA confirmed that the NCCN Guideline (v.2.2013) contains the following description regarding T-DM1 and that T-DM1 has not been described in representative text books on clinical oncology or in diagnosis and treatment guidelines in Japan.

- Based on the results of the EMILIA study, T-DM1 is recommended for treatment of patients with HER2-positive metastatic or recurrent breast cancer after treatment with trastuzumab.

The applicant explained the clinical positioning and target patients as follows:
In the EMILIA study, administration of T-DM1 to patients with HER2-positive inoperable or recurrent breast cancer who had previously been treated with chemotherapy with a taxane antineoplastic drug and trastuzumab significantly improved OS and PFS, parameters assessed as the primary endpoints, compared to that of lapatinib/Cape, one of the standard treatments after trastuzumab treatment. No significant differences were observed in the safety profile or PK between the Japanese and foreign patients, taking account of the results of the Japanese and foreign clinical studies. It is expected that T-DM1-associated risks will be controllable by appropriate measures such as close monitoring, dose reduction, and treatment interruption, and thus that T-DM1 will be well tolerated. Therefore, T-DM1 will be useful as a new standard treatment, in place of the conventional treatment with lapatinib/Cape, for patients with HER2-positive inoperable or recurrent breast cancer who have previously been treated with chemotherapy with a taxane antineoplastic drug and trastuzumab, the target patient group in the EMILIA study.

The efficacy and safety of T-DM1 have been demonstrated only in patients with a history of chemotherapy with a taxane antineoplastic drug and trastuzumab. For other patients, no sufficient evidence has been available to recommend T-DM1 at the moment.

[redacted]

[redacted].

Based on the above, the applicant explained that it would be appropriate to set the indication of T-DM1 as "HER2-positive inoperable or recurrent breast cancer" and that caution should be provided in the Precautions for Indications section that T-DM1 should be administered to patients who have received prior treatment with trastuzumab and a taxane antineoplastic drug.

Based on the results of the review in "4.(iii).B.(2) Efficacy" and "4.(iii).B.(3) Safety," PMDA concluded that T-DM1 may be positioned as a treatment option for patients with HER2-positive inoperable or recurrent breast cancer who have been treated with chemotherapy with a taxane antineoplastic drug and trastuzumab, and accepted the applicant's explanation regarding Indications and Precautions for Indications.

4.(iii).B.(4).2) Selection of patients based on HER2 expression level

PMDA asked the applicant to explain the setting of the inclusion criterion related to HER2 expression level in the submitted clinical studies and the efficacy by HER2 level in the EMILIA study.

The applicant responded as follows:
In all submitted clinical studies, the definition of HER-2 positive was "IHC3+ or FISH-positive."

OS outcome by HER2 expression level in the EMILIA study was as shown in the following table. It indicates that the hazard ratios [95% CI] of the T-DM1 group to the lapatinib/Cape group in the HER2 positive subpopulations were 0.61 [0.44, 0.83] in patients determined as FISH-positive and IHC 3+; 0.60 [0.29, 1.26] in patients as FISH-positive and IHC 2+; and 0.67 [0.28, 1.56] in patients as FISH unknown and IHC 3+, showing that the hazard ratios were consistent with that observed in the entire population. There were only a limited number of patients in subpopulations of FISH-positive and IHC 0; FISH-positive and IHC 1+; FISH positive and IHC unknown; and FISH-negative and IHC 3+, precluding the evaluation of OS. Thus, although evaluation was difficult in some subpopulations, there was no information suggesting a clear difference in efficacy among "IHC 3+ or FISH-positive" patients who were enrolled in the EMILIA study as HER2-positive patients. Therefore, T-DM1 is recommended to "IHC 3+ or FISH-positive" patients.

HER2 expression level and OS outcome (EMILIA study)

	No. of patients enrolled	T-DM1 group			Lapatinib/Cape group			Hazard ratio [95% CI]
		No. of patients	No. of events	Median OS (months)	No. of patients	No. of events	Median OS (months)	
All patients	991	495	94	NE	496	129	23.3	0.63 [0.48, 0.82]
FISH-positive and IHC 3+	758	365	63	27.1	393	100	23.7	0.61 [0.44, 0.83]
FISH-positive and IHC 2+	104	56	13	NE	48	15	23.2	0.60 [0.29, 1.26]
FISH-unknown and IHC 3+	89	53	13	NE	36	9	22.6	0.67 [0.28, 1.56]
FISH-positive and IHC 0/1+	11	7	4	15.2	4	1	NE	2.12 [0.24, 19.11]
FISH-positive and IHC unknown	6	5	0	NE	1	1	21.9	NE [NE, NE]
FISH-negative and IHC 3+	8	2	0	NE	6	0	NE	NE [NE, NE]

NE: not evaluable

PMDA considers as follows:
There are only limited data available for the subpopulations of IHC unknown/0/1+ and FISH-positive as well as IHC3+ and FISH-negative among IHC3+ or FIH-positive patients, the patient population proposed for T-DM1 treatment. Therefore, it is practically impossible to derive any conclusion on efficacy in these subpopulations. However, PMDA accepted the applicant's response that T-DM1 is recommended for "IHC3+ or FISH-positive" patients, by taking account of the results of the review in "Review Report on Perjeta Intravenous Infusion 420 mg/14 mL dated April 9, 2013." However, it is necessary to provide information, using information materials, etc., to facilitate understanding the criteria for the target patients for T-DM1 and trastuzumab therapy, based on HER2 expression level (IHC 3+ or FISH-positive).

4.(iii).B.(4).3) Efficacy and safety as neoadjuvant or adjuvant chemotherapy
Taking account of the fact that efficacy or safety of T-DM1 in neoadjuvant or adjuvant chemotherapy has not been demonstrated in clinical studies at the moment, the applicant explained that caution will be provided regarding this provision in the Precautions for Indications section of the package insert.

PMDA accepted the applicant's explanation.

4.(iii).B.(5) Dosage and administration
The proposed dosage and administration was "The usual adult dosage is 3.6 mg/kg (body weight) of Trastuzumab Emtansine given as an intravenous infusion every 3 weeks."

As a result of the following review, PMDA has concluded that the dosage and administration should be modified as "The usual adult dosage is 3.6 mg/kg (body weight) of Trastuzumab Emtansine (Genetical Recombination) given as an intravenous infusion every 3 weeks by intravenous infusion." PMDA has also concluded that it is appropriate to provide the following cautions in the Precautions for Dosage and Administration section of the package insert.

- If administration has been postponed for any reason, T-DM1 should be administered as soon as the treatment can be resumed followed by dosing every 3 weeks.

- Criteria for treatment interruption, dose reduction, or treatment discontinuation depending on the symptoms and severity of adverse drug reactions.

- The initial dose should be administered over 90 minutes. If the initial dose is well tolerated, the duration of administration for the second and subsequent doses can be shortened to a minimum of 30 minutes.

- The efficacy and safety of combination therapy with other antineoplastic drugs have not been established.

4.(iii).B.(5).1) Dose and dosing intervals
The applicant explained the justification for the proposed dosage and administration as follows:
In the foreign phase I study (Study TDM3569g), 6 dose levels (0.3, 0.6, 1.2, 2.4, 3.6, 4.8 mg/kg) of T-DM1 were investigated by every-3-week dosing, and 5 dose levels (1.2, 1.6, 2.0, 2.4, 2.9 mg/kg) of T-DM1 were investigated by every week dosing. As a result, MTD was determined to be 3.6 mg/kg for every-3-week dosing and 2.4 mg/kg for every week dosing. Based on these results, the EMILIA study was conducted using the dosage regimen of 3.6 mg/kg every 3 weeks. OS and PFS significantly improved in the T-DM1 group compared with the lapatinib/Cape group, and the tolerability of T-DM1 was confirmed.

In the Japanese phase I study (Study JO22591), the dosage regimens of T-DM1 1.8, 2.4, and 3.6

mg/kg every 3 weeks were investigated. The probability of DLT estimated by CRM was closest to 25% at the dose of 3.6 mg/kg, from which MTD was estimated to be 3.6 mg/kg. Based on these results, the Japanese phase II study (Study JO22997) was conducted using the dosage regimen of T-DM1 3.6 mg/kg every 3 weeks. As a result, the response rate was similar to that observed in foreign phase II studies (Studies TDM4258g and TDM4374g) which were conducted in the same target population as in Study JO22997, and the tolerability of T-DM1 was confirmed.

Based on the above results, the dosage and administration were proposed as "T-DM1 3.6 mg/kg every 3 weeks."

PMDA accepted the applicant's explanation and concluded it is appropriate to set the dosage and administration as proposed.

4.(iii).B.(5).2) Criteria for treatment interruption, dose reduction, and treatment discontinuation

The applicant explained the criteria for treatment interruption, dose reduction, and treatment discontinuation of T-DM1 as follows:
In the EMILIA study and Study JO22997, the following criteria for treatment interruption, dose reduction, and treatment discontinuation of T-DM1 were specified. Since it was considered possible to ensure the tolerability of T-DM1 by complying with these criteria, almost the same criteria were included in the Precautions for Dosage and Administration section of the package insert (b and c below have been changed from the criteria in the EMILIA study and Study JO22997 [see below]).

In the EMILIA study, when T-DM1 treatment was postponed for any reason such as adverse events, it was not necessary to postpone the treatment until the scheduled date in the next cycle; instead, the treatment was allowed to be resumed as soon as possible and subsequent treatment was performed every 3 weeks. Therefore, such information should be included in the Precautions for Dosage and Administration section to raise cautions.

Criteria for treatment interruption, dose reduction, and treatment discontinuation
In the first dose reduction, approximately 80% of the usual dose (3.0 mg/kg [body weight]) should be used. In the second dose reduction, appropriately 80% of the first reduced dose (2.4 mg/kg [body weight]) should be used. If further reduction is necessary, administration should be discontinued.

a. Criteria for treatment interruption or treatment discontinuation on the basis of decreased left ventricular ejection fraction (LVEF)

b. Criteria for treatment interruption, dose reduction, or treatment discontinuation on the basis of increased AST or ALT:
Hepatotoxicity was observed as an adverse event characteristic of T-DM1. Therefore, if AST increased or ALT increased was Grade 4 or met Hy's law case (AST or ALT $>3 \times$ ULN and total bilirubin $>2 \times$ ULN), administration should be discontinued, although these discontinuation criteria were not included in the EMILIA study or Study JO22997.

c. Criteria for treatment interruption, dose reduction, or treatment discontinuation on the basis of hyperbilirubinaemia:
The EMILIA study or Study JO22997 specified that administration should be withdrawn temporarily if a Grade ≥ 2 hyperbilirubinaemia occurred and, after improving to Grade ≤ 1, administration could be resumed at 1 level below the previous dose. However, results of clinical studies so far obtained showed that Grade 2 bilirubinaemia was often accompanied

by AST increased or ALT increased. Therefore, it was specified that, when only bilirubin increased up to Grade 2, administration could be resumed after recovery without dose reduction. Also, since hepatotoxicity was observed as an adverse event characteristic to T-DM1, administration was to be discontinued if bilirubinaemia was Grade 4 or met Hy's law case (AST or ALT >3 × ULN and total bilirubin >2 × ULN) although these discontinuation criteria had not been included in the EMILIA study or Study JO22997.

d. Criteria for treatment interruption or dose reduction on the basis of thrombocytopenia:
The EMILIA study specified that administration should be withdrawn temporarily if a Grade ≥4 thrombocytopenia occurred and, after thrombocytopenia improved to Grade ≤1, administration could be resumed at 1 level below the previous dose. In contrast, Study JO22997 specified that T-DM1 could be administered if thrombocytopenia was Grade ≤1 or baseline. In either study, T-DM1 has never been administered to patients with Grade ≥3 thrombocytopenia. Therefore, T-DM1 is to be temporarily withdrawn if Grade ≥3 thrombocytopenia occurs.

e. Criteria for treatment interruption on the basis of peripheral neuropathy

PMDA accepted the applicant's explanation.

4.(iii).B.(5).3) Infusion speed

The applicant explained the infusion speed of T-DM1 as follows:
Since T-DM1 is an antibody product, infusion reaction may occur during administration. In clinical studies on T-DM1, the initial dose was to be administered over 90 minutes and, if the dose was well-tolerated, the duration of administration for the second and subsequent doses could be shortened to a minimum of 30 minutes. The reduction of the infusion time in the second and subsequent doses did not have any clear effect on the occurrence of adverse events such as infusion reaction. Therefore, caution will be provided in the Precautions for Dosages and Administration section of the package insert that the initial dose should be administered over 90 minutes and, if the initial dose is well-tolerated, the duration of administration for the second and subsequent doses can be shortened to a minimum of 30 minutes.

PMDA accepted the applicant's explanation.

4.(iii).B.(5).4) Concomitant use with other antineoplastic drugs

The applicant explained as follows:
The efficacy and safety of T-DM1 were confirmed when T-DM1 was administered as a monotherapy at 3.6 mg/kg, but not when concomitantly administered with other antineoplastic drugs. This information will be provided in the Precautions for Dosage and Administration section of the package insert to raise caution.

PMDA accepted the applicant's explanation.

4.(iii).B.(6) Post-marketing investigations

The applicant explained the post-marketing investigations as follows:
The applicant plans to conduct a post-marketing surveillance, using the central registration system, in patients with HER2-positive inoperable or recurrent breast cancer treated with T-DM1 in order to identify the safety of the T-DM1 under routine use of T-DM1 after the market launch.

The priority investigation item was platelet count decreased because, based on the Japanese and foreign clinical studies, Grade ≥3 platelet count decreased occurred with a higher incidence in Japanese patients than in foreign patients and because platelet count decreased caused treatment

interruption at the highest frequency among adverse events in the Japanese clinical studies.

In the Japanese phase II study (Study JO22997), T-DM1 treatment was postponed because of platelet count decreased in 17.8% (13 of 73 patients) of patients (mostly due to the low baseline level, at Grade ≥2). Assuming that the incidence of platelet count decreased (Grade ≥2) before T-DM1 treatment under routine use of the drug is similar to the percentage of patients who postponed the treatment due to platelet count decreased in Study JO22997 (15%-20%), the incidence of platelet count decreased and the margin from the lower limit of 95% CI may be estimated with the probability of ≤0.05 (5%) if data are accumulated from 228 patients. Based on the above, the target sample size in this post-marketing surveillance was set at 250 patients to allow for possible withdrawals from the surveillance.

The recruitment period of the post-marketing surveillance was set at 9 months. Since platelet count decreased was observed frequently during the early stage of treatment in both Japanese and foreign clinical studies, it is expected that sufficient information on the incidence of platelet count decreased will be collected before the end of Cycle 8 (6 months). However, another objective of the post-marketing surveillance is to collect information on the efficacy such as PFS. Therefore, by taking account of the median PFS of 9.6 months in the EMILIA study, the observation period was set at 1 year.

PMDA considers as follows:
In the EMILIA study, the incidences of Grade ≥3 adverse events and serious adverse events in the T-DM1 group did not show any higher tendency compared with the lapatinib/Cape group, when all types of adverse events were combined as a whole. In contrast, the incidences of Grade ≥3 thrombocytopenia and hepatotoxicity tended to be higher in T-DM1 group. Also, comparison of the incidence of Grade ≥3 adverse events between the EMILIA study and Study JO22997 showed that adverse events such as thrombocytopenia and hepatotoxicity occurred more frequently in Japanese patients, requiring particular caution in the administration of T-DM1. In addition, given the limited number of Japanese patients who have been treated with T-DM1, it is necessary to conduct a post-marketing surveillance to collect safety information under the routine use in Japan.

In addition to platelet count decreased proposed by the applicant as the priority investigation item, the objectives of the post-marketing surveillance should include hepatotoxicity because Grade ≥3 hepatotoxicity occurred with a higher incidence in Japanese patients compared with foreign patients. The target sample size and the duration of the observation period should be re-examined in line with the change of the priority investigation items.

4.(iv) Adverse events, etc. observed in clinical studies

Deaths reported in clinical studies submitted as the safety evaluation data are described in "4.(iii) Summary of clinical efficacy and safety." Major adverse events other than deaths were shown below.

4.(iv).(1) Japanese phase I study (Study JO22591)

Adverse events were observed in all the patients (100%) in the 1.8 mg/kg, the 2.4 mg/kg, and the 3.6 mg/kg groups. Adverse events for which a causal relationship to T-DM1 could not be ruled out were also observed in all the patients. Adverse events with an incidence of ≥20% in any treatment group were shown in the following table.

Adverse events with an incidence of ≥20%

Event	Number of patients					
	1.8 mg/kg group N = 1		2.4 mg/kg group N = 4		3.6 mg/kg group N = 5	
	All Grades	Grade ≥3	All Grades	Grade ≥3	All Grades	Grade ≥3
All adverse events	1 (100)	0	4 (100)	4 (100)	5 (100)	0
Nausea	1 (100)	0	2 (50.0)	0	4 (80.0)	0
Arthralgia	1 (100)	0	3 (75.0)	0	3 (60.0)	0
Pyrexia	1 (100)	0	2 (50.0)	0	3 (60.0)	0
Fatigue	1 (100)	0	1 (25.0)	0	3 (60.0)	0
Decreased appetite	0	0	1 (25.0)	0	4 (80.0)	0
Diarrhoea	0	0	1 (25.0)	0	3 (60.0)	0
Malaise	0	0	1 (25.0)	0	3 (60.0)	0
Nasopharyngitis	0	0	3 (75.0)	0	1 (20.0)	0
Headache	1 (100)	0	1 (25.0)	0	2 (40.0)	0
Rash	0	0	2 (50.0)	0	2 (40.0)	0
Constipation	0	0	1 (25.0)	0	2 (40.0)	0
Vomiting	0	0	0	0	3 (60.0)	0
Myalgia	0	0	1 (25.0)	0	2 (40.0)	0
Chills	1 (100)	0	2 (50.0)	0	0	0
Cystitis	0	0	1 (25.0)	0	2 (40.0)	0
Gastritis	0	0	2 (50.0)	0	0	0
Gingivitis	0	0	1 (25.0)	0	1 (20.0)	0
Musculoskeletal pain	0	0	1 (25.0)	0	1 (20.0)	0
Pain in extremity	0	0	2 (50.0)	0	0	0
Oedema peripheral	0	0	1 (25.0)	0	1 (20.0)	0
AST increased	0	0	2 (50.0)	1 (25.0)	0	0
ALT increased	0	0	2 (50.0)	1 (25.0)	0	0
Dyspnoea	0	0	1 (25.0)	0	1 (20.0)	0
Oropharyngeal pain	0	0	1 (25.0)	0	1 (20.0)	0
Epistaxis	0	0	1 (25.0)	0	1 (20.0)	0
Rhinorrhoea	0	0	1 (25.0)	0	1 (20.0)	0
Pericardial effusion	0	0	0	0	2 (40.0)	0
Blood ALP increased	0	0	1 (25.0)	0	1 (20.0)	0

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, ALP: Alkaline phosphatase

Serious adverse events were observed in 1 of 4 patients (25.0%) in the 2.4 mg/kg group and in 1 of 5 patients (20.0%) in the 3.6 mg/kg group, and were cholelithiasis and gastric ulcer haemorrhage, respectively. A causal relationship to T-DM1 could not be ruled out for both events.

Adverse events leading to discontinuation of T-DM1 were reported by 2 of 4 patients (50.0%) in the 2.4 mg/kg group and 1 of 5 patients (20.0%) in the 3.6 mg/kg group. These were thrombocytopenia and cholelithiasis (1 patient each) in the 2.4 mg/kg group and blood ALP increased (1 patient) in the 3.6 mg/kg group. A causal relationship to T-DM1 could not be ruled out for any of the events.

4.(iv).(2) Japanese phase II study (Study JO22997)

Adverse events were observed in 70 of 73 patients (95.9%), and adverse events for which a causal relationship to T-DM1 could not be ruled out were observed in 67 of 73 patients (91.8%). Adverse events with an incidence of ≥10% were as shown in the following table.

Adverse events with incidence of ≥10%

Event	Number of patients (%)	
	T-DM1 group N = 73	
	All Grades	Grade ≥3
All adverse events	70 (95.9)	41 (56.2)
Nausea	32 (43.8)	0
Epistaxis	30 (41.1)	0
Pyrexia	26 (35.6)	0
Nasopharyngitis	24 (32.9)	0
Malaise	23 (31.5)	0
Decreased appetite	22 (30.1)	2 (2.7)
Platelet count decreased	20 (27.4)	16 (21.9)
AST increased	15 (20.5)	10 (13.7)
Headache	14 (19.2)	0
Vomiting	14 (19.2)	4 (5.5)
Stomatitis	12 (16.4)	0
Constipation	12 (16.4)	0
Neutrophil count decreased	10 (13.7)	2 (2.7)
Rash	10 (13.7)	0
Fatigue	10 (13.7)	1 (1.4)
ALT increased	8 (11.0)	6 (8.2)
Chills	8 (11.0)	0
Cough	8 (11.0)	0
Peripheral sensory neuropathy	8 (11.0)	1 (1.4)

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase

Serious adverse events were observed in 15 of 73 patients (20.5%). The serious adverse events observed were malaise (2 patients [2.7%]), pyrexia, implant site erythema, upper limb fracture, humerus fracture, femoral neck fracture, cerebral haemorrhage, brain oedema, peripheral sensory neuropathy, acute respiratory distress syndrome, pulmonary embolism, oesophageal ulcer, pneumonia, hepatic function abnormal, retinal detachment, pathological fracture, stress cardiomyopathy, mental status changes, and decreased appetite (1 patient each [1.4%]). Of these, a causal relationship to T-DM1 could not be ruled out for malaise (2 patients), pyrexia, cerebral haemorrhage, brain oedema, peripheral sensory neuropathy, acute respiratory distress syndrome, pulmonary embolism, hepatic function abnormal, stress cardiomyopathy, or decreased appetite (1 patient each).

Adverse events leading to discontinuation of T-DM1 were reported by 14 of 73 patients (19.2%). These were AST increased, haemoglobin decreased, γ–GTP increased, ejection fraction decreased, platelet count decreased, blood ALP increased, blood bilirubin increased, neutrophil count decreased, weight decreased, brain oedema, peripheral sensory neuropathy, oedema peripheral, pathological fracture, anaemia, acute respiratory distress syndrome, femoral neck fracture, hypoalbuminaemia, and skin ulcer (1 patient each [1.4%]). Of these, a causal relationship to T-DM1 could not be ruled out for AST increased, haemoglobin decreased, γ–GTP increased, ejection fraction decreased, platelet count decreased, blood ALP increased, blood bilirubin increased, neutrophil count decreased, weight decreased, brain oedema, peripheral sensory neuropathy, acute respiratory distress syndrome, or skin ulcer (1 patient each).

4.(iv).(3) Foreign phase I study (Study TDM3569g)

4.(iv).(3).1) Every-3-week dosing

Adverse events were observed in all the patients (100%) in the 0.3 mg/kg, the 0.6 mg/kg, the 1.2 mg/kg, the 2.4 mg/kg, the 3.6 mg/kg, and the 4.8 mg/kg groups. Adverse events for which a causal relationship to T-DM1 could not be ruled out were observed in 2 of 3 patients (66.7%) in the 0.3 mg/kg group, 1 of 1 patient (100%) in the 0.6 mg/kg group, 1 of 1 patient (100%) in the 1.2 mg/kg group, 1 of 1 patient (100%) in the 2.4 mg/kg group, 14 of 15 patients (93.3%) in the 3.6 mg/kg

group, and 3 of 3 patients (100%) in the 4.8 mg/kg group. Adverse events with an incidence of ≥20% in all groups combined receiving every-3-week dosing were as shown in the following table.

Adverse events with an incidence of ≥20% in all groups combined receiving every-3-week dosing

Event	Number of patients (%)							
	0.3 mg/kg group N = 3		0.6 mg/kg group N = 1		1.2 mg/kg group N = 1		2.4 mg/kg group N = 1	
	All Grades	Grade ≥3	All Grades	Grade ≥3	All Grades	Grade 3	All Grades	≥Grade 3
All adverse events	3 (100)	1 (33.3)	1 (100)	1 (100)	1 (100)	0	1 (100)	0
Thrombocytopenia	0	0	0	0	0	0	1 (100)	0
Fatigue	1 (33.3)	0	0	0	0	0	1 (100)	0
Nausea	2 (66.7)	0	1 (100)	0	0	0	1 (100)	0
Anaemia	0	0	0	0	1 (100)	0	0	0
Constipation	2 (66.7)	0	0	0	0	0	1 (100)	0
Cough	1 (33.3)	0	1 (100)	0	0	0	0	0
Headache	0	0	0	0	0	0	1 (100)	0
Arthralgia	1 (33.3)	0	0	0	0	0	1 (100)	0
Back pain	0	0	0	0	0	0	1 (100)	0
Contusion	0	0	0	0	0	0	1 (100)	0
Dyspnoea	2 (66.7)	1 (33.3)	0	0	0	0	0	0
Urinary tract infection	0	0	0	0	0	0	0	0
Inappetence	2 (66.7)	0	0	0	1 (100)	0	0	0
AST increased	0	0	0	0	0	0	1 (100)	0
Dry mouth	0	0	0	0	1 (100)	0	1 (100)	0
Epistaxis	1 (33.3)	0	0	0	0	0	0	0
Hypokalaemia	0	0	0	0	0	0	0	0
Upper respiratory tract infection	0	0	0	0	0	0	1 (100)	0

AST: Aspartate aminotransferase

Adverse events with an incidence of ≥20% in all groups combined receiving every-3-week dosing (continued)

Event	Number of patients (%)					
	3.6 mg/kg group N = 15		4.8 mg/kg group N = 3		Sum of every-3-week dosing groups N = 24	
	All Grades	Grade ≥3	All Grades	Grade ≥3	All Grades	Grade ≥3
All adverse events	15 (100)	7 (46.7)	3 (100)	3 (100)	24 (100)	12 (50.0)
Thrombocytopenia	9 (60.0)	1 (33.3)	3 (100)	2 (66.7)	13 (54.2)	3 (12.5)
Fatigue	9 (60.0)	0	1 (33.3)	0	12 (50.0)	0
Nausea	6 (40.0)	0	1 (33.3)	0	11 (45.8)	0
Anaemia	6 (40.0)	0	0	0	7 (29.2)	0
Constipation	3 (20.0)	0	1 (33.3)	0	7 (29.2)	0
Cough	4 (26.7)	0	1 (33.3)	0	7 (29.2)	0
Headache	5 (33.3)	0	1 (33.3)	0	7 (29.2)	0
Arthralgia	3 (20.0)	0	1 (33.3)	0	6 (25.0)	0
Back pain	4 (26.7)	0	1 (33.3)	0	6 (25.0)	0
Contusion	5 (33.3)	0	0	0	6 (25.0)	0
Dyspnoea	4 (26.7)	0	0	0	6 (25.0)	1 (4.2)
Urinary tract infection	5 (33.3)	0	1 (33.3)	0	6 (25.0)	0
Inappetence	2 (13.3)	0	0	0	5 (20.8)	0
AST increased	2 (13.3)	0	2 (66.7)	0	5 (20.8)	0
Dry mouth	3 (20.0)	0	0	0	5 (20.8)	0
Epistaxis	3 (20.0)	0	1 (33.3)	0	5 (20.8)	0
Hypokalaemia	4 (26.7)	0	1 (33.3)	0	5 (20.8)	0
Upper respiratory tract infection	4 (26.7)	0	0	0	5 (20.8)	0

AST: Aspartate aminotransferase

Serious adverse events were observed in 1 of 3 patients (33.3%) in the 0.3 mg/kg group, 1 of 1 patient (100%) in the 0.6 mg/kg group, 3 of 15 patients (20.0%) in the 3.6 mg/kg group, and in 2 of 3 patients (66.7%) in the 4.8 mg/kg group. These were dyspnoea (1 patient) in the 0.3 mg/kg group, pleural effusion (1 patient) in the 0.6 mg/kg group, cerebral haemorrhage, humerus fracture, pulmonary hypertension, cellulitis, convulsion, and brain oedema (1 patient each) in the 3.6 mg/kg group, and convulsion and dysarthria in 1 patient each in the 4.8 mg/kg group. Of these, a causal relationship to T-DM1 could not be ruled out for pulmonary hypertension (1 patient) in the 3.6 mg/kg group.

Adverse events leading to discontinuation of T-DM1 were reported by 2 of 15 patients (13.3%) in the 3.6 mg/kg group and 2 of 3 patients (66.7%) in the 4.8 mg group. These were muscular weakness and pulmonary hypertension (1 patient each) in the 3.6 mg/kg group and thrombocytopenia (2 patients) in the 4.8 mg/kg group. Of these, a causal relationship to T-DM1 could not be ruled out for pulmonary hypertension (1 patient) in the 3.6 mg/kg group or thrombocytopenia (2 patients) in the 4.8 mg/kg group.

4.(iv).(3).2) Every week dosing

Adverse events were observed in all patients in the 1.2 mg/kg, the 1.6 mg/kg, the 2.0 mg/kg, the 2.4 mg/kg, and the 2.9 mg/kg groups. Adverse events for which a causal relationship to T-DM1 could not be ruled out were observed in 3 of 3 patients (100%) in the 1.2 mg/kg group, 2 of 3 patients (66.7%) in the 1.6 mg/kg group, 3 of 3 patients (100%) in the 2.0 mg/kg group, 14 of 16 patients (87.5%) in the 2.4 mg/kg group, and 3 of 3 patients (100%) in the 2.9 mg/kg group. Adverse events with an incidence of ≥20% in all groups combined receiving every week dosing were as shown in the following table.

Adverse events with an incidence of ≥20% in all groups combined receiving every week dosing

Event	Number of patients (%)					
	1.2 mg/kg N = 3		1.6 mg/kg N = 3		2.0 mg/kg N = 3	
	All Grades	Grade ≥3	All Grades	Grade ≥3	All Grades	Grade ≥3
All adverse events	3 (100)	1 (33.3)	3 (100)	2 (66.7)	3 (100)	2 (66.7)
Fatigue	3 (100)	0	1 (33.3)	0	3 (100)	1 (33.3)
Nausea	3 (100)	0	1 (33.3)	0	1 (33.3)	0
AST increased	1 (33.3)	1 (33.3)	1 (33.3)	1 (33.3)	2 (66.7)	0
Diarrhoea	3 (100)	0	1 (33.3)	0	2 (66.7)	0
Thrombocytopenia	1 (33.3)	0	1 (33.3)	1 (33.3)	2 (66.7)	0
Headache	2 (66.7)	0	1 (33.3)	0	1 (33.3)	0
Dry mouth	1 (33.3)	0	1 (33.3)	0	1 (33.3)	0
Dyspnoea	1 (33.3)	0	0	0	0	0
Constipation	1 (33.3)	0	0	0	2 (66.7)	0
Cough	1 (33.3)	0	1 (33.3)	0	2 (66.7)	0
Vomiting	2 (66.7)	0	1 (33.3)	0	0	0
Anaemia	0	0	0	0	1 (33.3)	0
Epistaxis	0	0	1 (33.3)	0	2 (66.7)	0
Pyrexia	2 (66.7)	0	1 (33.3)	0	2 (66.7)	0
Inappetence	2 (66.7)	0	0	0	0	0
Insomnia	2 (66.7)	0	0	0	2 (66.7)	0
Hypokalaemia	2 (66.7)	1 (33.3)	1 (33.3)	0	1 (33.3)	0

AST: Aspartate aminotransferase

Adverse events with an incidence of ≥20% in all groups combined receiving every week dosing (continued)

Event	Number of patients (%)					
	2.4 mg/kg N = 16		2.9 mg/kg N = 3		Sum of every week dosing groups N = 28	
	All Grades	Grade ≥3	All Grades	Grade ≥3	All Grades	Grade ≥3
All adverse events	16 (100)	13 (81.3)	3 (100)	1 (33.3)	28 (100)	19 (67.9)
Fatigue	10 (62.5)	1 (6.3)	1 (33.3)	0	18 (64.3)	2 (7.1)
Nausea	7 (43.8)	0	2 (66.7)	0	14 (50.0)	0
AST increased	8 (50.0)	1 (6.3)	1 (33.3)	0	13 (46.4)	3 (10.7)
Diarrhoea	6 (37.5)	0	1 (33.3)	0	13 (46.4)	0
Thrombocytopenia	6 (37.5)	1 (6.3)	2 (66.7)	1 (33.3)	12 (42.9)	3 (10.7)
Headache	7 (43.8)	0	0	0	11 (39.3)	0
Dry mouth	7 (43.8)	0	0	0	10 (35.7)	0
Dyspnoea	9 (56.3)	2 (12.5)	0	0	10 (35.7)	2 (7.1)
Constipation	4 (25.0)	0	2 (66.7)	0	9 (32.1)	0
Cough	5 (31.3)	0	0	0	9 (32.1)	0
Vomiting	6 (37.5)	0	0	0	9 (32.1)	0
Anaemia	6 (37.5)	4 (25.0)	1 (33.3)	0	8 (28.6)	4 (14.3)
Epistaxis	5 (31.3)	0	0	0	8 (28.6)	0
Pyrexia	2 (12.5)	0	1 (33.3)	0	8 (28.6)	0
Inappetence	4 (25.0)	0	1 (33.3)	0	7 (25.0)	0
Insomnia	3 (18.8)	0	0	0	7 (25.0)	0
Hypokalaemia	2 (12.5)	1 (6.3)	0	0	6 (21.4)	2 (7.1)

AST: Aspartate aminotransferase

Serious adverse events were observed in 1 of 3 patients (33.3%) in the 1.2 mg/kg group, 1 of 3 patients (33.3%) in the 1.6 mg/kg group, 1 of 3 patients (33.3%) in the 2.0 mg/kg group, and 8 of 16 patients (50.0%) in the 2.4 mg/kg group. These were confusional state (1 patient) in the 1.2 mg/kg group; pneumonia (1 patient) in the 1.6 mg/kg group; pneumonitis (1 patient) in the 2.0 mg/kg group; pneumonia (2 patients), and osteomyelitis, dyspnoea, pain, influenza, hepatic encephalopathy, pulmonary embolism, and cellulitis (1 patient each) in the 2.4 mg/kg group. Of these, a causal relationship to T-DM1 could not be ruled out for pneumonitis (1 patient) in the 2.0 mg/kg group.

Adverse events leading to discontinuation of T-DM1 were reported by 3 of 16 patients (18.8%) in the 2.4 mg/kg group and 1 of 3 patients (33.3%) in the 2.9 mg/kg group. These were pain, fatigue, and dyspnoea (1 patient each) in the 2.4 mg/kg group and thrombocytopenia (1 patient) in the 2.9 mg/kg group. Of these, a causal relationship to T-DM1 could not be ruled out for fatigue, dyspnoea (1 patient each) in the 2.4 mg/kg group or thrombocytopenia (1 patient) in the 2.9 mg/kg group.

4.(iv).(4) Foreign phase II study (Study TDM4258g)

Adverse events were observed in all the patients (100%). Adverse events for which a causal relationship to T-DM1 could not be ruled out were observed in 102 of 112 patients (91.1%). Adverse events with an incidence of ≥10% were as shown in the following table.

Adverse events with an incidence of ≥10%

Event	Number of patients (%) T-DM1 group N = 112	
	All Grades	Grade ≥3
All adverse events	112 (100)	50 (44.6)
Fatigue	73 (65.2)	5 (4.5)
Nausea	57 (50.9)	1 (0.9)
Headache	45 (40.2)	0
Epistaxis	40 (35.7)	2 (1.8)
Pyrexia	39 (34.8)	1 (0.9)
Constipation	34 (30.4)	0
Cough	31 (27.7)	0
Diarrhoea	29 (25.9)	0
Hypokalaemia	27 (24.1)	10 (8.9)
Vomiting	27 (24.1)	1 (0.9)
Arthralgia	25 (22.3)	1 (0.9)
Pain in extremity	25 (22.3)	0
Dyspnoea	23 (20.5)	3 (2.7)
Anaemia	23 (20.5)	3 (2.7)
Chills	22 (19.6)	0
Inappetence	22 (19.6)	1 (0.9)
Upper respiratory tract infection	20 (17.9)	0
Neuropathy peripheral	20 (17.9)	1 (0.9)
Thrombocytopenia	19 (17.0)	9 (8.0)
Rash	19 (17.0)	0
Urinary tract infection	18 (16.1)	0
Dry mouth	16 (14.3)	0
Decreased appetite	16 (14.3)	0
Insomnia	15 (13.4)	0
Depression	14 (12.5)	0
Back pain	14 (12.5)	1 (0.9)
Musculoskeletal pain	13 (11.6)	0
Weight decreased	13 (11.6)	0
Dizziness	13 (11.6)	1 (0.9)
Pain	13 (11.6)	1 (0.9)
Contusion	12 (10.7)	0
Dyspepsia	12 (10.7)	0
Anxiety	12 (10.7)	0
Abdominal pain	12 (10.7)	0
Oedema peripheral	12 (10.7)	0

Serious adverse events were observed in 30 of 112 patients (26.8%). These were cellulitis (3 patients [2.7%]), pneumonia, back pain, convulsion, confusional state, dyspnoea, and pleural effusion (2 patients each [1.8%]), and osteomyelitis, urosepsis, pneumothorax, respiratory failure, arthralgia, groin pain, musculoskeletal chest pain, altered state of consciousness, aphasia, cerebrovascular accident, haemorrhoidal haemorrhage, dysphagia, oesophageal stenosis, upper gastrointestinal haemorrhage, hip fracture, subdural haemorrhage, asthenia, disease progression, dehydration, failure to thrive, hepatic enzyme increased, platelet count decreased, white blood cell count increased, hepatotoxicity, thrombocytopenia, deep vein thrombosis, and acute myeloid leukaemia (1 patient each [0.9%]). Of these, a causal relationship to T-DM1 could not be ruled out for cellulitis, haemorrhoidal haemorrhage, dehydration, hepatic enzyme increased, platelet count decreased, hepatotoxicity, or thrombocytopenia (1 patient each).

Adverse events leading to discontinuation of T-DM1 were reported by 4 of 112 patients (3.6%). These were platelet count decreased (2 patients [1.8%]), and hepatic enzyme increased, asthenia, failure to thrive, and acute myeloid leukaemia (1 patient each [0.9%]). Of these, a causal relationship to T-DM1 could not be ruled out for platelet count decreased (2 patients) or hepatic

enzyme increased (1 patient).

4.(iv).(5) Foreign phase II study (Study TDM4374g)

Adverse events were observed in all the patients (100%). Adverse events for which a causal relationship to T-DM1 could not be ruled out were observed in 97 of 110 patients (88.2%). Adverse events with an incidence of ≥10% were as shown in the following table.

Adverse events with an incidence of ≥10%

Event	Number of patients (%) T-DM1 group N = 110	
	All Grades	Grade ≥3
All adverse events	110 (100)	54 (49.1)
Fatigue	69 (62.7)	5 (4.5)
Nausea	41 (37.3)	1 (0.9)
Thrombocytopenia	37 (33.6)	8 (7.3)
AST increased	30 (27.3)	3 (2.7)
Constipation	27 (24.5)	1 (0.9)
Headache	26 (23.6)	1 (0.9)
Pyrexia	26 (23.6)	1 (0.9)
Hypokalaemia	25 (22.7)	1 (0.9)
Epistaxis	25 (22.7)	1 (0.9)
Decreased appetite	23 (20.9)	1 (0.9)
Anaemia	23 (20.9)	2 (1.8)
Dry mouth	22 (20.0)	0
Cough	21 (19.1)	0
Back pain	20 (18.2)	3 (2.7)
Neuropathy peripheral	20 (18.2)	0
Dyspnoea	18 (16.4)	3 (2.7)
Vomiting	18 (16.4)	0
ALT increased	16 (14.5)	3 (2.7)
Arthralgia	16 (14.5)	2 (1.8)
Pain in extremity	16 (14.5)	1 (0.9)
Myalgia	15 (13.6)	0
Infusion related reaction	15 (13.6)	0
Diarrhoea	14 (12.7)	0
Muscle spasms	13 (11.8)	0
Blood ALP increased	12 (10.9)	0
Weight decreased	12 (10.9)	1 (0.9)
Depression	11 (10.0)	3 (2.7)
Chills	11 (10.0)	0
Urinary tract infection	11 (10.0)	0
Abdominal pain	11 (10.0)	2 (1.8)
Oedema peripheral	11 (10.0)	0

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, ALP: Alkaline phosphatase

Serious adverse events were observed in 29 of 110 patients (26.4%). These were cellulitis (4 patients [3.6%]), pneumonia and pyrexia (3 patients each [2.7%]), nausea, axillary pain, sepsis, spinal cord compression, and dyspnoea (2 patients each [1.8%]), and bacteraemia, gastroenteritis viral, herpes zoster, pharyngitis streptococcal, staphylococcal infection, wound infection, chest pain, influenza like illness, vomiting, abdominal pain, abdominal pain upper, constipation, pancreatitis, interstitial lung disease, pleural effusion, pneumothorax, fall, femur fracture, foreign body, road traffic accident, central nervous system necrosis, convulsion, hepatic function abnormal, bile duct obstruction, cholecystitis acute, blood creatinine increased, blood culture positive, deep vein thrombosis, atrial fibrillation, vaginal haemorrhage, confusional state, and fluid overload (1 patient each [0.9%]). Of these, a causal relationship to T-DM1 could not be ruled out for nausea, pneumonia, pyrexia, vomiting, hepatic function abnormal, or atrial fibrillation (1 patient each).

Adverse events leading to discontinuation of T-DM1 were reported by 7 of 110 patients (6.4%). These were hepatic function abnormal, cholelithiasis, pancreatitis, fatigue, thrombocytopenia, dyspnoea, atrial fibrillation, and spinal cord compression (1 patient each [0.9%]). Of these, a causal relationship to T-DM1 could not be ruled out for hepatic function abnormal, fatigue, thrombocytopenia, or atrial fibrillation (1 patient each).

4.(iv).(6) Foreign phase III study (Study TDM4370g/BO21977)

Adverse events were observed in 470 of 490 patients (95.9%) in the T-DM1 group and in 477 of 488 patients (97.7%) in the lapatinib/Cape group. Adverse events for which a causal relationship to T-DM1 could not be ruled out were observed in 427 of 490 patients (87.1%), and adverse events for which a causal relationship to lapatinib/Cape could not be ruled out in 468 of 488 patients (95.9%). Adverse events with an incidence of ≥10% in the T-DM1 group were as shown in the following table.

Adverse events with an incidence of ≥10% in the T-DM1 group

Event	Number of patients (%)			
	T-DM1 group N = 490		Lapatinib/Cape group N = 488	
	All Grades	Grade ≥3	All Grades	Grade ≥3
Adverse events	470 (95.9)	200 (40.8)	477 (97.7)	278 (57.0)
Nausea	192 (39.2)	4 (0.8)	218 (44.7)	12 (2.5)
Fatigue	172 (35.1)	12 (2.4)	136 (27.9)	17 (3.5)
Thrombocytopenia	137 (28.0)	63 (12.9)	12 (2.5)	1 (0.2)
Headache	133 (27.1)	4 (0.8)	68 (13.9)	4 (0.8)
Constipation	124 (25.3)	2 (0.4)	47 (9.6)	0
Diarrhoea	114 (23.3)	8 (1.6)	389 (79.7)	101 (20.7)
AST increased	110 (22.4)	21 (4.3)	46 (9.4)	4 (0.8)
Decreased appetite	101 (20.6)	2 (0.4)	113 (23.2)	5 (1.0)
Epistaxis	99 (20.2)	1 (0.2)	39 (8.0)	0
Vomiting	93 (19.0)	4 (0.8)	143 (29.3)	22 (4.5)
Asthenia	86 (17.6)	2 (0.4)	81 (16.6)	7 (1.4)
Arthralgia	85 (17.3)	3 (0.6)	38 (7.8)	0
Pyrexia	85 (17.3)	0	37 (7.6)	2 (0.4)
ALT increased	83 (16.9)	14 (2.9)	43 (8.8)	7 (1.4)
Cough	83 (16.9)	1 (0.2)	60 (12.3)	1 (0.2)
Dry mouth	77 (15.7)	0	24 (4.9)	1 (0.2)
Myalgia	69 (14.1)	3 (0.6)	18 (3.7)	0
Back pain	64 (13.1)	3 (0.6)	50 (10.2)	2 (0.4)
Abdominal pain upper	57 (11.6)	2 (0.4)	41 (8.4)	1 (0.2)
Dyspnoea	56 (11.4)	3 (0.6)	36 (7.4)	2 (0.4)
Insomnia	54 (11.0)	2 (0.4)	41 (8.4)	1 (0.2)
Pain in extremity	52 (10.6)	2 (0.4)	52 (10.7)	5 (1.0)
Rash	52 (10.6)	0	130 (26.6)	9 (1.8)
Anaemia	51 (10.4)	13 (2.7)	39 (8.0)	8 (1.6)
Neuropathy peripheral	49 (10.0)	8 (1.6)	28 (5.7)	1 (0.2)

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase

Serious adverse events were observed in 76 of 490 patients (15.5%) in the T-DM1 group and in 88 of 488 patients (18.0%) in the lapatinib/Cape group. Serious adverse events observed in the T-DM1 group were pyrexia (7 patients [1.4%]), vomiting (6 patients [1.2%]), abdominal pain (4 patients [0.8%]), urinary tract infection and thrombocytopenia (3 patients each [0.6%]), diarrhoea, gastrointestinal haemorrhage, cellulitis, device related infection, sepsis, pain, chest pain, femur fracture, back pain, hyponatraemia, and metrorrhagia (2 patients each [0.4%]), and nausea, colitis, constipation, gastric ulcer, gastritis, gastrointestinal obstruction, bacteraemia, clostridium difficile

colitis, tooth infection, upper respiratory tract infection, appendicitis, bronchitis, enterococcal infection, gastroenteritis norovirus, herpes zoster, nasopharyngitis, parotitis, pneumonia, pneumonia bacterial, staphylococcal sepsis, injection site extravasation, fatigue, acute respiratory distress syndrome, pleural effusion, pneumonitis, haemoptysis, hypoxia, dizziness, headache, cerebrovascular accident, metabolic encephalopathy, Parkinson's disease, status epilepticus, infusion related reaction, ankle fracture, haemolytic transfusion reaction, hip fracture, open wound, wrist fracture, neutropenia, atrial fibrillation, cardiomyopathy, pericarditis, supraventricular tachycardia, bone pain, muscular weakness, pathological fracture, spinal column stenosis, spondylitis, failure to thrive, hypokalaemia, hepatitis toxic, hepatotoxicity, portal hypertension, cholangitis, dermatitis contact, skin haemorrhage, confusional state, dysuria, renal failure, colon cancer, myelodysplastic syndrome, platelet transfusion, hypersensitivity, blood bilirubin increased, and γ–GTP increased (1 patient each [0.2%]). Serious adverse events observed in the lapatinib/Cape group were diarrhoea (17 patients [3.5%]), vomiting (9 patients [1.8%]), pulmonary embolism (7 patients [1.4%]), nausea, cellulitis, and pyrexia (3 patients each [0.6%]), abdominal pain, ileus, bacteraemia, acute respiratory distress syndrome, dizziness, headache, femur fracture, febrile neutropenia, pericardial effusion, dehydration, and deep vein thrombosis (2 patients each [0.4%]), and enteritis, intestinal obstruction, ileal fistula, peptic ulcer haemorrhage, device related infection, sepsis, clostridium difficile colitis, lower respiratory tract infection, salmonellosis, erysipelas, H1N1 influenza, pyelonephritis acute, pain, asthenia, mucosal inflammation, multi-organ failure, oedema peripheral, pleural effusion, alveolitis allergic, asthma, pleuritic pain, pulmonary oedema, respiratory failure, cerebrovascular accident, syncope, coma, hydrocephalus, extradural haematoma, fall, femoral neck fracture, subdural haemorrhage, thrombocytopenia, neutropenia, anaemia of malignant disease, angina pectoris, coronary artery disease, pain in extremity, hyponatraemia, hyperbilirubinaemia, thrombosis, jugular vein thrombosis, subclavian vein thrombosis, venous thrombosis, rash generalised, urticaria, scar, metrorrhagia, ovarian cyst, confusional state, agitation, depression, ureteric obstruction, urinary tract obstruction, and vertigo (1 patient each [0.2%]). Of these, a causal relationship to T-DM1 could not be ruled out for pyrexia (4 patients); vomiting or thrombocytopenia (3 patients each); chest pain (2 patients); or abdominal pain, nausea, gastrointestinal haemorrhage, cellulitis, urinary tract infection, tooth infection, upper respiratory tract infection, injection site extravasation, acute respiratory distress syndrome, pneumonitis, metabolic encephalopathy, Parkinson's disease, infusion related reaction, atrial fibrillation, cardiomyopathy, pericarditis, supraventricular tachycardia, failure to thrive, hepatitis toxic, hepatotoxicity, portal hypertension, platelet transfusion, hypersensitivity, blood bilirubin increased, or γ–GTP increased (1 patient each). A causal relationship to lapatinib or Cape could not be ruled out for diarrhoea (17 patients); vomiting (9 patients); nausea, abdominal pain or febrile neutropenia (2 patients each); or pulmonary embolism, cellulitis, ileus, dehydration, enteritis, intestinal obstruction, lower respiratory tract infection, salmonellosis, asthenia, mucosal inflammation, multi-organ failure, syncope, thrombocytopenia, neutropenia, anaemia of malignant disease, angina pectoris, coronary artery disease, hyponatraemia, thrombosis, rash generalised, urticaria, or agitation (1 patient each).

Adverse events leading to discontinuation of T-DM1 or lapatinib/Cape were reported by 29 of 490 patients (5.9%) in the T-DM1 group and 52 of 488 patients (10.7%) in the lapatinib/Cape group. Adverse events leading to discontinuation of T-DM1 were thrombocytopenia (10 patients [2.0%]), AST increased (4 patients [0.8%]), blood bilirubin increased and hyperbilirubinaemia (2 patients each [0.4%]), and leukopenia, anaemia of chronic disease, dyspnoea, acute respiratory distress syndrome, cough, pneumonitis, lethargy, neuropathy peripheral, ALT increased, hepatitis toxic, hepatotoxicity, bronchitis, left ventricular dysfunction, and infusion related reaction (1 patient each [0.2%]). Adverse events leading to discontinuation of lapatinib/Cape were diarrhoea (16 patients [3.3%]), vomiting (12 patients [2.5%]), palmar-plantar erythrodysaesthesia syndrome (10 patients [2.0%]), nausea (3 patients [0.6%]), abdominal pain, stomatitis, leukopenia, dyspnoea, fatigue, and paronychia (2 patients each [0.4%]), and

gastritis, anaemia, neutropenia, erythema, pulmonary embolism, ejection fraction decreased, dizziness, sensory disturbance, hydrocephalus, mucosal inflammation, angina pectoris, pain in extremity, venous thrombosis, agitation, and dehydration (1 patient each [0.2%]). Of these, a causal relationship to T-DM1 could not be ruled out for thrombocytopenia (10 patients); AST increased (4 patients); blood bilirubin increased or hyperbilirubinaemia (2 patients each); or leukopenia, anaemia of chronic disease, dyspnoea, acute respiratory distress syndrome, cough, pneumonitis, lethargy, neuropathy peripheral, ALT increased, hepatitis toxic, hepatotoxicity, left ventricular dysfunction, or infusion related reaction (1 patient each). A causal relationship to lapatinib or Cape could not be ruled out for diarrhoea (16 patients); vomiting (12 patients); palmar-plantar erythrodysaesthesia syndrome (10 patients); nausea (3 patients); abdominal pain, stomatitis, leukopenia, dyspnoea, fatigue, or paronychia (2 patients each); or gastritis, anaemia, neutropenia, erythema, dizziness, sensory disturbance, mucosal inflammation, angina pectoris, agitation, or dehydration (1 patient each).

4.(iv).(7) Foreign phase I/II study (Study TDM4373g/BO22495)

Adverse events were observed in all the patients (100%) in the 3.0 mg and the 3.6 mg groups in the phase I part and in all the patients (100%) in the 3.6 mg group in the phase II part. Adverse events for which a causal relationship to T-DM1 or pertuzumab could not be ruled out were observed in 3 of 3 patients (100%) in the 3.0 mg group and 6 of 6 patients (100%) in the 3.6 mg group in the phase I part, and in 56 of 58 patients (96.6%) in the phase II part. Adverse events with an incidence of ≥20% in all the treatment groups combined were as shown in the following table.

Adverse events with an incidence of ≥20% in all treatment groups combined

Event	Number of patients (%)							
	Phase I part				Phase II part		All groups combined N = 67	
	3.0 mg/kg group N = 3		3.6 mg/kg group N = 6		3.6 mg/kg group N = 58			
	All Grades	Grade ≥3	All Grades	Grade ≥3	All Grades	Grade ≥3	All Grades	Grade ≥3
All adverse events	3 (100)	1 (33.3)	6 (100)	2 (33.3)	58 (100)	35 (60.3)	67 (100)	38 (56.7)
Fatigue	3 (100)	1 (33.3)	4 (66.7)	2 (33.3)	35 (60.3)	5 (8.6)	42 (62.7)	8 (11.9)
Nausea	2 (66.7)	0	3 (50.0)	0	29 (50.0)	2 (3.4)	34 (50.7)	2 (3.0)
Diarrhoea	1 (33.3)	0	3 (50.0)	0	22 (37.9)	1 (1.7)	26 (38.8)	1 (1.5)
Cough	1 (33.3)	0	2 (33.3)	0	22 (37.9)	0	25 (37.3)	0
Decreased appetite	2 (66.7)	0	1 (16.7)	0	21 (36.2)	2 (3.4)	24 (35.8)	2 (3.0)
Thrombocytopenia	0	0	1 (16.7)	1 (16.7)	20 (34.5)	7 (12.1)	21 (31.3)	8 (11.9)
Constipation	1 (33.3)	0	1 (16.7)	0	20 (34.5)	0	22 (32.8)	0
Chills	2 (66.7)	0	1 (16.7)	0	19 (32.8)	0	22 (32.8)	0
Pyrexia	1 (33.3)	0	2 (33.3)	0	18 (31.0)	0	21 (31.3)	0
AST increased	0	0	2 (33.3)	0	17 (29.3)	6 (10.3)	19 (28.4)	6 (9.0)
Vomiting	0	0	1 (16.7)	0	18 (31.0)	2 (3.4)	19 (28.4)	2 (3.0)
ALT increased	0	0	2 (33.3)	0	16 (27.6)	6 (10.3)	18 (26.9)	6 (9.0)
Dyspnoea	2 (66.7)	0	2 (33.3)	0	15 (25.9)	4 (6.9)	19 (28.4)	4 (6.0)
Headache	2 (66.7)	0	3 (50.0)	0	13	0	18 (26.9)	0

Event	Number of patients (%)							
	Phase I part				Phase II part		All groups combined N = 67	
	3.0 mg/kg group N = 3		3.6 mg/kg group N = 6		3.6 mg/kg group N = 58			
	All Grades	Grade ≥3	All Grades	Grade ≥3	All Grades	Grade ≥3	All Grades	Grade ≥3
					(22.4)			
Mucosal inflammation	0	0	2 (33.3)	0	14 (24.1)	0	16 (23.9)	0
Epistaxis	2 (66.7)	0	2 (33.3)	0	14 (24.1)	0	18 (26.9)	0
Peripheral sensory neuropathy	1 (33.3)	0	2 (33.3)	0	14 (24.1)	2 (3.4)	17 (25.4)	2 (3.0)
Dysgeusia	2 (66.7)	0	3 (50.0)	0	13 (22.4)	0	18 (26.9)	0
Rash	1 (33.3)	0	1 (16.7)	0	14 (24.1)	1 (1.7)	16 (23.9)	1 (1.5)
Insomnia	0	0	2 (33.3)	0	13 (22.4)	0	15 (22.4)	0
Arthralgia	1 (33.3)	0	2 (33.3)	0	11 (19.0)	0	14 (20.9)	0

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase

Serious adverse events were observed in 1 of 6 patients (16.7%) in the 3.6 mg group in the phase I part and in 21 of 58 patients (36.2%) in the 3.6 mg group in the phase II part. Serious adverse events observed in the 3.6 mg group in the phase I part were pneumonia and pleural effusion (1 patient each [16.7%]). Serious adverse events observed in the 3.6 mg group in the phase II part were cellulitis and dyspnoea (3 patients each [5.2%]), abdominal pain, nausea, pleural effusion, pneumonia, and vomiting (2 patients each [3.4%]), and pyrexia, pain, aphasia, tachycardia, failure to thrive, breast cellulitis, localised infection, osteomyelitis, skin infection, staphylococcal bacteraemia, urinary tract infection, diarrhoea, ileus, colitis, gastritis, pneumonitis, fatigue, ataxia, cerebral haemorrhage, pericardial effusion, haematuria, renal failure acute, diabetic foot, hepatic cirrhosis, and tumour haemorrhage (1 patient each [1.7%]). Of these, a causal relationship to T-DM1 or pertuzumab could not be ruled out for pleural effusion (1 patient) in the 3.6 mg/kg group in the phase I part; or vomiting (2 patients), abdominal pain, pyrexia, tachycardia, nausea, diarrhoea, pneumonitis, ileus, fatigue, haematuria, or hepatic cirrhosis (1 patient each) in the 3.6 mg group in the phase II part.

Adverse events leading to discontinuation of T-DM1 or pertuzumab were reported by 1 of 3 patients (33.3%) in the 3.0 mg group in the phase I part and 10 of 58 patients (17.2%) in the 3.6 mg group in the phase II part. These were fatigue in 1 patient (33.3%) in the 3.0 mg group in the phase I part; and fatigue in 3 patients (5.2%), aphasia, epistaxis, gingival bleeding, rectal haemorrhage, AST increased, neuralgia, cerebral haemorrhage, chills, pyrexia, haemoptysis, hepatic cirrhosis, left ventricular dysfunction, and erythema in 1 patient each (1.7%) in the 3.6 mg group in the phase II part. Of these, a causal relationship to T-DM1 or pertuzumab could not be ruled out for fatigue (1 patient) in the 3.0 mg in the phase I part; or fatigue (3 patients), epistaxis, gingival bleeding, rectal haemorrhage, AST increased, neuralgia, chills, pyrexia, hepatic cirrhosis, left ventricular dysfunction, or erythema (1 patient each) in the 3.6 mg in the phase II part.

4.(iv).(8) Foreign phase II study (Study TDM4450g/BO21976)

Adverse events were observed in 66 of 69 patients (95.7%) in the T-DM1 group and in 66 of 66 patients (100%) in the trastuzumab/DTX group. Adverse events for which a causal relationship to T-DM1 could not be ruled out were observed in 63 of 69 patients (91.3%), and adverse events for which a causal relationship to trastuzumab or DTX could not be ruled out in 66 of 66 patients (100%). Adverse events with an incidence of ≥10% in the T-DM1 group were as shown in the

following table.

Adverse events with an incidence of ≥10% in T-DM1 group

Event	Number of patients (%)			
	T-DM1 group N = 69		Trastuzumab/DTX group N = 66	
	All Grades	Grade ≥3	All Grades	Grade ≥3
All adverse events	66 (95.7)	32 (46.4)	66 (100)	60 (90.9)
Nausea	34 (49.3)	2 (2.9)	29 (43.9)	0
Fatigue	34 (49.3)	3 (4.3)	30 (45.5)	3 (4.5)
AST increased	30 (43.5)	6 (8.7)	4 (6.1)	0
Headache	28 (40.6)	0	12 (18.2)	0
Pyrexia	28 (40.6)	0	15 (22.7)	1 (1.5)
Thrombocytopenia	19 (27.5)	5 (7.2)	4 (6.1)	2 (3.0)
Back pain	19 (27.5)	1 (1.4)	21 (31.8)	3 (4.5)
Epistaxis	19 (27.5)	0	6 (9.1)	0
ALT increased	18 (26.1)	7 (10.1)	4 (6.1)	0
Cough	18 (26.1)	0	14 (21.2)	0
Vomiting	17 (24.6)	2 (2.9)	17 (25.8)	0
Arthralgia	16 (23.2)	0	20 (30.3)	1 (1.5)
Constipation	16 (23.2)	1 (1.4)	15 (22.7)	0
Asthenia	16 (23.2)	2 (2.9)	14 (21.2)	2 (3.0)
Decreased appetite	13 (18.8)	1 (1.4)	11 (16.7)	0
Abdominal pain upper	12 (17.4)	0	4 (6.1)	0
Hypokalaemia	12 (17.4)	2 (2.9)	6 (9.1)	0
Rash	12 (17.4)	0	15 (22.7)	0
Chills	11 (15.9)	0	5 (7.6)	0
Diarrhoea	11 (15.9)	0	30 (45.5)	2 (3.0)
Dry mouth	11 (15.9)	0	3 (4.5)	0
Neutropenia	11 (15.9)	4 (5.8)	43 (65.2)	41 (62.1)
Dyspepsia	11 (15.9)	0	8 (12.1)	0
Nasopharyngitis	11 (15.9)	0	10 (15.2)	0
Blood ALP increased	10 (14.5)	2 (2.9)	2 (3.0)	0
Dyspnoea	10 (14.5)	0	18 (27.3)	2 (3.0)
Upper respiratory tract infection	10 (14.5)	0	8 (12.1)	0
Neuropathy peripheral	10 (14.5)	0	10 (15.2)	1 (1.5)
Musculoskeletal pain	9 (13.0)	0	4 (6.1)	0
Hypertension	9 (13.0)	1 (1.4)	6 (9.1)	0
Pain in extremity	9 (13.0)	1 (1.4)	15 (22.7)	1 (1.5)
Anaemia	9 (13.0)	2 (2.9)	18 (27.3)	3 (4.5)
Oropharyngeal pain	8 (11.6)	0	8 (12.1)	0
Stomatitis	8 (11.6)	0	13 (19.7)	0
Infusion related reaction	8 (11.6)	0	4 (6.1)	0
Insomnia	8 (11.6)	0	12 (18.2)	1 (1.5)
Myalgia	7 (10.1)	0	12 (18.2)	0
Nail disorder	7 (10.1)	0	16 (24.2)	0
Leukopenia	7 (10.1)	0	17 (25.8)	16 (24.2)
Sinusitis	7 (10.1)	0	5 (7.6)	0
Abdominal pain	7 (10.1)	1 (1.4)	4 (6.1)	0
Oedema peripheral	7 (10.1)	0	29 (43.9)	4 (6.1)

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, ALP: Alkaline phosphatase

Serious adverse events were observed in 14 of 69 patients (20.3%) in the T-DM1 group and in 17 of 66 patients (25.8%) in the trastuzumab/DTX group. Serious adverse events observed in the T-DM1 group were pneumonia (5 patients [7.2%]), sepsis, pleural effusion, pneumonitis, pleuritic pain, chills, pyrexia, sudden death, tumour lysis syndrome, dehydration, hypercalcaemia, hypertensive crisis, hypovolaemic shock, atrial fibrillation, supraventricular extrasystoles, abdominal pain, and vomiting (1 patient each [1.4%]). Serious adverse events observed in the trastuzumab/DTX group were febrile neutropenia (6 patients [9.1%]), cellulitis (2 patients [3.0%]), arthritis infective, anaemia, pleural effusion, alveolitis allergic, dyspnoea, epistaxis, pulmonary

embolism, oedema peripheral, hyperglycaemia, metabolic acidosis, deep vein thrombosis, atrial fibrillation, cardiopulmonary failure, intestinal obstruction, spinal compression fracture, renal failure acute, hypersensitivity, breast cancer, and C-reactive protein increased (1 patient each [1.5%]). Of these, a causal relationship to T-DM1 could not be ruled out for pneumonitis, chills, tumour lysis syndrome, hypertensive crisis, hypovolaemic shock, atrial fibrillation, supraventricular extrasystoles, abdominal pain, or vomiting (1 patient each). A causal relationship to trastuzumab or DTX could not be ruled out for febrile neutropenia (6 patients), pleural effusion, alveolitis allergic, oedema peripheral, or hypersensitivity (1 patient each).

Adverse events leading to discontinuation of T-DM1 were reported by 5 of 69 patients (7.2%) and that of trastuzumab or DTX by 23 of 66 patients (34.8%). These were ALT increased and AST increased (3 patients each [4.3%]), and thrombocytopenia and neuropathy peripheral (1 patient each [1.4%]) in the T-DM1 group; and peripheral sensory neuropathy (4 patients [6.1%]), neuropathy peripheral and oedema peripheral (3 patients each [4.5%]), face oedema, fatigue, alveolitis allergic, pneumonitis, dyspnoea, onychomadesis, palmar-plantar erythrodysaesthesia syndrome, skin reaction, diarrhoea, neutropenia, cardiopulmonary failure, hypersensitivity, breast cancer, and ejection fraction decreased (1 patient each [1.5%]) in the trastuzumab/DTX group. Of these, a causal relationship to T-DM1 could not be ruled out for ALT increased and AST increased (2 patients each), and thrombocytopenia and neuropathy peripheral (1 patient each). A causal relationship to trastuzumab or DTX could not be ruled out for peripheral sensory neuropathy (4 patients); neuropathy peripheral or oedema peripheral (3 patients each); face oedema, fatigue, alveolitis allergic, pneumonitis, onychomadesis, palmar-plantar erythrodysaesthesia syndrome, skin reaction, diarrhoea, neutropenia, hypersensitivity, or ejection fraction decreased (1 patient each).

4.(iv).(9) Foreign phase II study (Study TDM4688g)

Adverse events were observed in 51 of 51 patients (100%) in the T-DM1 monotherapy period and in 18 of 20 patients (90.0%) in the T-DM1/pertuzumab treatment period. Adverse events for which a causal relationship to T-DM1 could not be ruled out were observed in 50 of 51 patients (98.0%). Adverse events for which a causal relationship to T-DM1 or pertuzumab could not be ruled out in 13 of 20 patients (65.0%). Adverse events with an incidence of ≥10% in either period were as shown in the following table.

Adverse events with an incidence of ≥10% in either period

Event	T-DM1 monotherapy period N = 51		T-DM1/pertuzumab period N = 20	
	All Grades	Grade≥ 3	All Grades	Grade ≥3
All adverse events	51 (100)	17 (33.3)	18 (90.0)	12 (60.0)
Nausea	33 (64.7)	1 (2.0)	3 (15.0)	0
Fatigue	33 (64.7)	0	4 (20.0)	1 (5.0)
Dry mouth	25 (49.0)	0	2 (10.0)	0
Thrombocytopenia	18 (35.3)	4 (7.8)	2 (10.0)	1 (5.0)
AST increased	17 (33.3)	5 (9.8)	4 (20.0)	2 (10.0)
Vomiting	16 (31.4)	0	4 (20.0)	0
Decreased appetite	12 (23.5)	0	3 (15.0)	0
Hypokalaemia	12 (23.5)	0	3 (15.0)	2 (10.0)
Pyrexia	12 (23.5)	0	2 (10.0)	0
Constipation	12 (23.5)	0	1 (5.0)	0
Chills	11 (21.6)	0	0	0
Headache	11 (21.6)	0	2 (10.0)	0
Epistaxis	10 (19.6)	0	3 (15.0)	0
Arthralgia	9 (17.6)	1 (2.0)	1 (5.0)	0
Upper respiratory tract infection	9 (17.6)	0	3 (15.0)	0
Urinary tract infection	9 (17.6)	0	0	0
Cough	8 (15.7)	0	2 (10.0)	0
Blood lactate dehydrogenase increased	8 (15.7)	1 (2.0)	0	0
Dyspnoea	8 (15.7)	0	1 (5.0)	0
Insomnia	8 (15.7)	0	2 (10.0)	0
ALT increased	7 (13.7)	0	2 (10.0)	0
Hypertension	7 (13.7)	0	0	0
Anaemia	7 (13.7)	1 (2.0)	6 (30.0)	2 (10.0)
Musculoskeletal pain	6 (11.8)	0	2 (10.0)	0
Proteinuria	6 (11.8)	1 (2.0)	0	0
Neuropathy peripheral	6 (11.8)	1 (2.0)	1 (5.0)	0
Dysgeusia	6 (11.8)	0	0	0
Vision blurred	6 (11.8)	0	1 (5.0)	0
Weight decreased	5 (9.8)	0	2 (10.0)	0
Dizziness	5 (9.8)	0	2 (10.0)	0
Diarrhoea	4 (7.8)	0	3 (15.0)	1 (5.0)
Blood ALP increased	4 (7.8)	0	4 (20.0)	0
Back pain	4 (7.8)	1 (2.0)	2 (10.0)	0
Abdominal pain	4 (7.8)	0	2 (10.0)	0
Pain	4 (7.8)	0	4 (20.0)	0
Depression	3 (5.9)	1 (2.0)	3 (15.0)	1 (5.0)
Anxiety	3 (5.9)	0	2 (10.0)	0
Bone pain	2 (3.9)	1 (2.0)	3 (15.0)	1 (5.0)
Muscle spasms	1 (2.0)	0	2 (10.0)	0

AST: Aspartate aminotransferase, ALT: Alanine aminotransferase, ALP: Alkaline phosphatase

Serious adverse events were observed in 4 of 51 patients (7.8%) in the T-DM1 monotherapy period and in 5 of 20 patients (25.0%) in the T-DM1/pertuzumab treatment period. Serious adverse events observed in the T-DM1 monotherapy period were thrombocytopenia (2 patients [3.9%]), generalised oedema, dyspnoea, convulsion, and renal failure (1 patient each [2.0%]). Serious adverse events observed in the T-DM1/pertuzumab treatment period were anaemia (2 patients [10.0%]), thrombocytopenia, gastrointestinal haemorrhage, cardiac arrest, hypokalaemia, skin haemorrhage, and AST increased (1 patient each [5.0%]). Of these, a causal relationship to T-DM1 or pertuzumab could not be ruled out for thrombocytopenia (2 patients) in the T-DM1 monotherapy period; or thrombocytopenia or cardiac arrest (1 patient each) in the T-DM1/pertuzumab treatment period.

Adverse events leading to discontinuation of T-DM1 or pertuzumab reported by 2 of 51 patients (3.9%) in the T-DM1 monotherapy period and 5 of 20 patients (25.0%) in the T-DM1/pertuzumab

treatment period. These were ejection fraction decreased and hyperbilirubinaemia (1 patient each [2.0%]) in the T-DM1 monotherapy period and ejection fraction decreased, ALT increased, AST increased, blood ALP increased, gastrointestinal haemorrhage, oedema peripheral, and cardiac arrest (1 patient each [5.0%]) in the T-DM1/pertuzumab treatment period. Of these, a causal relationship to T-DM1 or pertuzumab could not be ruled out for ejection fraction decreased (1 patient) in the T-DM1 monotherapy period; or ejection fraction decreased, oedema peripheral, cardiac arrest (1 patient each) in the T-DM1/pertuzumab treatment period.

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

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

A document-based 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 product 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 (5.3.5.2-3). As a result, it was found that, in some clinical trial sites, blood samples for PK data were collected from subjects who had not given consent to participate in pharmacokinetic analysis. Thus, there were findings requiring improvements. However, since appropriate actions were taken for the findings, PMDA concluded that the clinical studies as a whole had been conducted in compliance with GCP and that there should be no problem with conducting a regulatory review based on the submitted product application documents.

IV. Overall Evaluation

Based on the submitted data, it is concluded that the efficacy of T-DM1 in patients with HER2-positive inoperable or recurrent breast cancer has been demonstrated and its safety is acceptable based on the observed clinical benefits. T-DM1 is a drug with a new active ingredient. It is considered that T-DM1, upon binding to HER2, induces antibody-dependent cell-mediated cytotoxicity, etc., as is the case with trastuzumab (genetical recombination) and then DM1, thus incorporated into cells, inhibits tumor growth by inducing cell cycle arrest and apoptosis. Thus, T-DM1 has a clinical significance as one of the options for the treatment of HER2-positive inoperable or recurrent breast cancer. The proposed indication and items to be investigated after the market launch will be further discussed at the Expert Discussion.

PMDA considers that T-DM1 may be approved if it can be concluded that there are no particular problems based on comments from the Expert Discussion.

Review Report (2)

August 12, 2013

I. Product Submitted for Registration

[Brand name]	Kadcyla Intravenous Infusion 100 mg Kadcyla Intravenous Infusion 160 mg
[Non-proprietary name]	Trastuzumab Emtansine (Genetical Recombination)
[Applicant]	Chugai Pharmaceutical Co., Ltd.
[Date of application]	January 29, 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

As a result of the review described in the "4.(iii).B.(2) Efficacy" of the Review Report (1), PMDA concluded that the efficacy of Trastuzumab Emtansine (Genetical Recombination) (hereinafter referred to as trastuzumab emtansine or T-DM1) in patients with human epidermal growth factor receptor type 2 (HER2)-positive, metastatic or recurrent breast cancer was demonstrated. The conclusion was based on the following two findings from the foreign phase III study (Study TDM4370g/Study BO21977, [EMILIA study]) conducted in patients with HER2-positive, metastatic or recurrent breast cancer with a history of chemotherapy with a taxane antineoplastic drug and trastuzumab (genetical recombination) (trastuzumab), by using combination of lapatinib tosilate hydrate (lapatinib) and capecitabine (Cape) (lapatinib/Cape group) as the control: (i) T-DM1 was superior to lapatinib/Cape in progression-free survival (PFS) (assessed by an independent review facility); and (ii) results (*P* value) of the additional interim analysis showed that T-DM1 significantly improved the overall survival.

The above conclusion of PMDA was supported by the expert advisors at the Expert Discussion.

(2) Safety

As a result of the review described in the "4.(iii).B.(3) Safety" of the Review Report (1), PMDA determined that adverse events requiring attention during treatment with T-DM1 were hepatotoxicity, nodular regenerative hyperplasia, thrombocytopenia, infusion reaction, interstitial lung disease, cardiac function failed, and neuropathy peripheral.

PMDA concluded as follows:
Attention should be paid to the occurrence of the adverse events set forth above in using T-DM1. However, T-DM1 is tolerable for Japanese patients with HER2-positive metastatic or recurrent breast cancer with a history of chemotherapy with a taxane antineoplastic drug and trastuzumab, provided that monitoring and managing of adverse events as well as the treatment interruption, dose reduction, treatment discontinuation, etc., are performed in an appropriate manner by physicians with sufficient knowledge and experience of cancer chemotherapy.

The above conclusion of PMDA was supported by the expert advisors at the Expert Discussion. Also, the following comments were raised by the expert advisors:

- Frequency of prophylactic administration of antiemetics against nausea and vomiting, and incidence of nausea and vomiting in patients who received prophylactic antiemetics should be investigated.

- Images of nodular regenerative hyperplasia and the time of onset of thrombocytopenia are useful information for clinical practice. Therefore, such information should be provided by using information materials, etc.

- In the EMILIA study, patients were excluded from the study if they had developed Grade ≥3 infusion reaction following trastuzumab administration in prior treatment but those with Grade ≤2 events were allowed to receive T-DM1. Although the Contraindications section of the package insert proposed by the applicant includes "patients with a history of hypersensitivity to trastuzumab," it is sometimes difficult to differentiate infusion reaction from hypersensitivity. Therefore, the details of Contraindications should be more clearly defined so that T-DM1 is not necessarily contraindicated for all patients, regardless of reaction grade, who have developed infusion reaction to trastuzumab in the prior treatment.

Taking account of the comments from the Expert Discussion, PMDA asked the applicant to explain the frequency of prophylactic administration of antiemetics against nausea and vomiting and the incidence of nausea and vomiting in patients who had received prophylactic antiemetics.

The applicant responded as follows:
In the T-DM1 group of the EMILIA study, prophylactic antiemetics were administered to 23 of 490 patients (4.7%). During T-DM1 infusion immediately after the administration of prophylactic antiemetics, Grade 2 vomiting occurred in 1 patient (4.3%) and Grade 1 nausea in 2 patients (8.7%) [Of a total of 490 patients in the T-DM1 group, 93 patients (19.0%) experienced vomiting of any grade and 4 (0.8%) had Grade ≥3 vomiting, and 192 (39.2%) experienced nausea of any grade and 4 (0.8%) had Grade ≥3 nausea: see Review Report (1) "4.(iii).B.(3).1 Safety profile and its difference between Japanese and foreign patients"]. In Study JO22997, prophylactic antiemetics were allowed only to patients who previously had nausea or vomiting to prevent the relapse; 12 of 73 patients (16.4%) met the requirement. During T-DM1 infusion immediately after the administration of prophylactic antiemetics, Grade 3 vomiting occurred in 2 patients (16.7%), Grade 1 nausea in 4 patients (33.3%), and Grade 2 nausea in 2 patients (16.7%) [Of a total of 73 patients in the T-DM1 group, 14 patients (19.2%) experienced vomiting of asny grade and 4 (5.5%) had Grade ≥3 vomiting, and 32 (43.8%) experienced nausea of any grade and none had Grade ≥3 nausea: see Review Report (1) "4.(iii).B.(3).1) Safety profile and its difference between Japanese and foreign patients"].

Based on the above results and the fact that the use of prophylactic antiemetics was not frequent either in the EMILIA study or Study JO22997, PMDA considers that there is no need to strongly recommend the use of prophylactic antiemetics. Instead, PMDA instructed the applicant to clarify the content of Contraindications and to provide the following information in clinical practice by using information materials, etc., to which the applicant agreed.

- Patients who developed Grade ≥3 infusion reaction after trastuzumab administration were excluded from the EMILIA study and Study JO22997.

- The frequency of prophylactic administration of antiemetics against nausea and vomiting and the incidence of nausea and vomiting in patients who received prophylactic antiemetics

- Examples of diagnostic images of nodular regenerative hyperplasia

- The time of onset of thrombocytopenia after administration of trastuzumab emtansine

(3) Clinical positioning and indications

As a result of the review described in the "4.(iii).B.(2) Efficacy" and "4.(iii).B.(3) Safety" of the Review Report (1), PMDA concluded that T-DM1 is positioned as an option for the treatment of patients with HER2-positive inoperable or recurrent breast cancer with a history of chemotherapy with a taxane antineoplastic drug and trastuzumab, and therefore that T-DM1 should be indicated for "HER2-positive inoperable or recurrent breast cancer," with the following statements in the Precautions for Indications section.

- HER2 positivity testing should be conducted by a pathologist or laboratory with sufficient experience.

- The efficacy and safety of trastuzumab emtansine in neoadjuvant or adjuvant chemotherapy have not been established.

- Trastuzumab emtansine should be administered to patients who have received prior treatment with trastuzumab and a taxane antineoplastic drug.

After having prepared the Review Report (1), PMDA confirmed that the most updated Japanese guideline for diagnosis and treatment of breast cancer "*Evidence-based Clinical Practice Guideline of Breast Cancer I. Treatment Methods*, 2013 ver., the Japanese Breast Cancer Society ed., Kanehara & Co., Ltd., 2013" states, based on the results of the EMILIA study, that T-DM1 is more effective than lapatinib/Cape in patients with HER2-positive metastatic or recurrent breast cancer with a history of chemotherapy with trastuzumab.

PMDA's above conclusion was supported by the expert advisors at the Expert Committee, with the following comments.

- Clinical development of T-DM1 was made covering a wide range of patients, such as those with a history of treatment with trastuzumab involved in Study JO22997. Therefore, it may be appropriate to exclude "patients who have received prior treatment with a taxane antineoplastic drug" from the Precautions for Indications section and include only "patients who have received prior treatment with trastuzumab."

Based on the above, PMDA instructed the applicant to set the Indications, and Precautions for Indications as follows, and the applicant accepted it.

Indication

HER2-positive inoperable or recurrent breast cancer

Precautions for Indications

- HER2 positivity testing should be conducted by a pathologist or laboratory with sufficient experience.

- The efficacy and safety of trastuzumab emtansine in neoadjuvant or adjuvant chemotherapy have not been established.

- Trastuzumab emtansine should be administered to patients who have received prior treatment with trastuzumab and a taxane antineoplastic drug.

(4) Dosage and administration

As a result of the review in "4.(iii).B.(5) Dosage and administration" of the Review Report (1),

the dosage and administration of T-DM1 should be set as follows: The usual adult dosage is 3.6 mg/kg (body weight) of Trastuzumab Emtansine (Genetical Recombination) given as an intravenous infusion every 3 weeks. PMDA also concluded that (a) to (d) below should be included in the Precautions for Dosage and Administration section of the package insert.

a. Criteria for treatment interruption, dose reduction, or treatment discontinuation depending on the symptoms and severity of adverse drug reactions (generally the same rules as those stipulated in the EMILIA study and Study JO22997) [see "4.(iii).B.(5).2) Criteria for treatment interruption, dose reduction, and treatment discontinuation" of the Review Report (1)]

b. If administration has been postponed for any reason, trastuzumab emtansine should be administered as soon as the treatment can be resumed, followed by dosing every 3 weeks.

c. The efficacy and safety of combination therapy with other antineoplastic drugs have not been established.

d. The initial dose should be administered over 90 minutes. If the initial dose is well tolerated, the duration of administration for the second and subsequent doses can be shortened to a minimum of 30 minutes.

The above conclusion of PMDA was generally supported by the expert advisors at the Expert Discussion. Also, a comment was made by some expert advisors that (b) above is a practice common to cancer chemotherapy and need not be included in the Precautions for Dosage and Administration section.

Based on the above, PMDA instructed the applicant to include the following Indications and Precautions for Dosage and Administration in the package insert, and the applicant accepted it.

Dosage and Administration
The usual adult dosage is 3.6 mg/kg (body weight) of Trastuzumab Emtansine (Genetical Recombination) given as an intravenous infusion every three weeks.

Precautions for Dosage and Administration
- The efficacy and safety of combination therapy with other antineoplastic drugs have not been established.

- The initial dose should be administered over 90 minutes. If the initial dose is well tolerated, the duration of administration for the second and subsequent doses can be shortened to a minimum of 30 minutes.

- Treatment interruption, dose reduction, or treatment discontinuation due to an adverse reaction should be decided based on its symptoms, severity, etc., taking account of the following criteria. The dose should not be increased once it is reduced.

Guide for dose reduction

Reduction Step	Dose Level
Starting dose	3.6 mg/kg
First dose reduction	3.0 mg/kg
Second dose reduction	2.4 mg/kg
Requirement for further dose reduction	Discontinue treatment.

(1) Criteria for treatment interruption or treatment discontinuation on the basis of decreased left ventricular ejection fraction (LVEF)

Adverse event		Measures
40% ≤ LVEF ≤ 45%	Absolute decrease is <10% points from baseline.	Continue trastuzumab emtansine treatment. Repeat LVEF assessment within 3 weeks.
	Absolute decrease is ≥10% points from baseline.	Do not administer trastuzumab emtansine treatment. Repeat LVEF assessment within 3 weeks. If LVEF has not recovered to within 10% points from baseline, discontinue trastuzumab emtansine.
LVEF <40%		Do not administer trastuzumab emtansine treatment. Repeat LVEF assessment within 3 weeks. If LVEF is still <40%, discontinue trastuzumab emtansine.
Symptomatic congestive cardiac failure		Discontinue trastuzumab emtansine.

(2) Criteria for treatment interruption, dose reduction, or treatment discontinuation on the basis of increased AST (GOT) or ALT (GPT)

Grade	Measures	
Grade 2 (>3 to 5 × ULN)	Treat at the same dose level.	* Discontinue trastuzumab emtansine if AST (GOT)/ALT (GPT) >3 × ULN and total bilirubin >2 × ULN.
Grade 3 (>5 to 20 × ULN)	Do not administer trastuzumab emtansine until AST (GOT)/ ALT (GPT) recovers to Grade ≤2, and then reduce one dose level.	
Grade 4 (>20 × ULN)	Discontinue trastuzumab emtansine.	

(3) Criteria for treatment interruption, dose reduction, or treatment discontinuation on the basis of hyperbilirubinaemia

Grade	Measures	
Grade 2 (>1.5 to 3 × ULN)	Do not administer trastuzumab emtansine until total bilirubin recovers to Grade ≤1, and then treat at the same dose level.	* Discontinue trastuzumab emtansine if AST (GOT)/ALT (GPT) >3 × ULN and total bilirubin >2 × ULN.
Grade 3 (>3 to 10 × ULN)	Do not administer trastuzumab emtansine until total bilirubin recovers to Grade ≤1, and then reduce one dose level.	
Grade 4 (>10 × ULN)	Discontinue trastuzumab emtansine.	

(4) Criteria for treatment interruption or dose reduction on the basis of thrombocytopenia

Grade	Measures
Grade 3 (25,000 to <50,000/mm^3)	Do not administer trastuzumab emtansine until platelet count recovers to Grade ≤1 (≥ 75,000/mm^3), and then treat at the same dose level.
Grade 4 (<25,000/mm^3)	Do not administer trastuzumab emtansine until platelet count recovers to Grade ≤1 (≥ 75,000/mm^3), and then reduce one dose level.

(5) Criteria for treatment interruption on the basis of peripheral neuropathy

Grade	Measures
Grade 3 or 4	Do not administer trastuzumab emtansine until symptoms recover to Grade ≤2, and then treat at the same dose level.

Grades are according to NCI CTCAE (v.4).
ULN: Upper limit of normal

- Trastuzumab emtansine is reconstituted with the Water for Injection (JP) included in the product package (5 mL for the 100-mg formulation and 8 mL for the 160-mg formulation) to obtain a trastuzumab emtansine (genetical recombination) solution at a concentration of

20 mg/mL. The required volume is then withdrawn from the vial using a syringe, immediately diluted into 250 mL of saline (JP), and intravenously infused.

(5) Risk management plan (draft)

In order to evaluate the safety, etc., of T-DM1 under routine use, the applicant plans to conduct a post-marketing surveillance of patients with HER2-positive inoperable or recurrent breast cancer treated with T-DM1 (250 patients, central registration system, 1-year follow-up period). The proposed priority investigation item for the post-marketing surveillance was platelet count decreased [see "4.(iii).B.(6) Post-marketing investigations" of the Review Report (1)].

As a result of its review in "4.(iii).B.(6) Post-marketing investigations" of the Review Report (1), PMDA concluded that a post-marketing surveillance should be conducted to collect safety information under routine use of T-DM1 in Japan. PMDA also concluded that priority investigation items for the surveillance should include, in addition to platelet count decreased proposed by the applicant, hepatotoxicity because Grade ≥3 events occurred more frequently in Japanese patients compared with foreign patients, and that the target sample size and the duration of the follow-up period should be re-examined in line with the change of the priority investigation items.

The above conclusion of PMDA was supported by the expert advisors at the Expert Discussion.

PMDA instructed the applicant to take appropriate measures accordingly.

The applicant responded as follows:
Hepatotoxicity will be included in the priority investigation items for the post-marketing surveillance in addition to platelet count decreased.

As regards the target sample size, attention was focused on treatment interruption, an event that affects treatment continuation. In Study JO22997, treatment interruption was mainly attributed to Grade ≥2 events observed before the administration. Thus, the target sample size for the post-marketing surveillance was to be set to confirm that the incidences of Grade ≥2 platelet count decreased and Grade ≥2 hepatotoxicity under routine use of T-DM1 are not significantly different from those observed in Study JO22997. Among patients in the study, treatment interruption occurred in 17.8% (13 of 73 patients) due to platelet count decreased and in 5.5% (4 of 73 patients) due to hepatotoxicity (aspartate aminotransferase increased, alanine aminotransferase increased). Therefore, assuming that the point estimate of each Grade ≥2 event under routine use of T-DM1 is similar to that the withdrawal rate in Study JO22997 (15-20% for platelet count decreased and 6% for hepatotoxicity), the incidences of platelet count decreased and hepatotoxicity and the margins from the lower limit of 95% CI may be estimated with the probability of ≤0.05 (5%) if data are accumulated from 228 patients and 42 patients, respectively. The target sample size will be set at 250 patients, taking also into account possible withdrawals from the post-marketing surveillance.

As regards the follow-up period, since hepatotoxicity was observed frequently during the early stage of the treatment in both Japanese and foreign clinical studies, it is expected that sufficient information on the occurrence of hepatotoxicity will be collected by the end of Cycle 8 (6 months). Therefore, the follow-up period will be set at 6 months.

PMDA accepted the applicant's response.

Based on the above discussion, PMDA has concluded, regarding the proposed risk management plan for T-DM1, that safety- and efficacy-related investigations should be carried out as shown in the following table and that additional pharmacovigilance activities and risk minimization

actions should be conducted.

Outline of drug risk management plan

Safety specifications		
Important identified risks	Important potential risks	Important missing information
• Interstitial lung disease (pneumonitis)/acute respiratory distress syndrome • Hepatic function disorder/nodular regenerative hyperplasia • Cardiac function failed (left ventricular dysfunction, cardiac failure congestive) • Hypersensitivity • Infusion reaction • Thrombocytopenia • Peripheral neuropathy	• None	• Safety in patients with hepatic impairment
Efficacy specifications		
• Efficacy under routine use of T-DM1		

Outline of pharmacovigilance plan and risk minimization plan

Additional pharmacovigilance activities	Additional risk minimization actions
• Early Post-marketing Phase Vigilance • Post-marketing surveillance (for outline, see the outline of use-results survey plan [draft] in the following table)	• Provision of information obtained from early post-marketing phase vigilance • Measures to prevent error in administration

Outline of use-results survey plan (draft)

Objective	To evaluate the safety, etc., of T-DM1 under routine use
Survey method	Central registration system
Patient population	Patients with HER2-positive, inoperable or recurrent breast cancer
Follow-up period	6 months
Planned sample size	250
Priority investigation items	Platelet count decreased, hepatic function disorder

III. Addition to Review Report (1)

The applicant's responses to the inquiries asked by PMDA at the time of preparation of the Review Report (1) are described below.

1. Data relating to quality

1.B. Outline of the review by PMDA

Shelf life of 100 mg formulation

The applicant submitted the data on 100-mg formulation that had been missing in the original submission, and explained that since the stability of the formulation up to 30 months was demonstrated by the additional data and by the data of long-term testing submitted previously, it is appropriate to propose the shelf life of 30 months for 100-mg formulation when stored at 5 ± 3°C.

PMDA accepted the applicant's explanation.

[redacted]

[redacted].

PMDA accepted the applicant's explanation.

IV. Overall Evaluation

As a result of the above review, PMDA concludes that the product may be approved after modifying the indication and the dosage and administration as shown below, with the conditions that appropriate cautions will be included in the package insert; information concerning the proper use of trastuzumab emtansine will be provided appropriately after the market launch; the compliance with the proper use of trastuzumab emtansine will be ensured under the supervision of a physician with sufficient knowledge and experience with cancer chemotherapy, and at a medical institution well equipped to cope with emergencies. The re-examination period is 8 years, the drug substance and the drug product are both classified as powerful drugs, and the product is classified as a biological product.

[Indication] HER2-positive inoperable or recurrent breast cancer

[Dosage and administration] The usual adult dosage is 3.6 mg/kg (body weight) of Trastuzumab Emtansine (Genetical Recombination) given as an intravenous infusion every 3 weeks by intravenous infusion.

[Warning]

1. Anticancer chemotherapy involving trastuzumab emtansine should be administered only to patients for whom trastuzumab emtansine is considered to be eligible, under the supervision of a physician with sufficient knowledge and experience with cancer chemotherapy, and at a medical institution well equipped to cope with emergencies. Eligible patients for trastuzumab emtansine should be selected with due care by referring to its package insert. Consent should be obtained, prior to the initiation of treatment, from the patient or his or her family member who has been provided with a thorough explanation of the benefits and risks of the therapy.

2. Cases of interstitial lung diseases (e.g., pneumonitis and interstitial pneumonia), some with fatal outcomes, have been reported with trastuzumab emtansine. Patients should therefore be carefully monitored by checking for initial symptoms (e.g., dyspnoea, cough, fatigue, pulmonary infiltration), performing chest X-ray, etc. If any abnormalities are found, appropriate measures, such as treatment discontinuation, should be taken.

[Contraindications]

1. Patients with a history of hypersensitivity (including serious, life-threatening infusion reactions difficult to distinguish from hypersensitivity) to trastuzumab (genetical recombination) or any ingredient in trastuzumab emtansine

2. Pregnant women or women who may be pregnant

[Precautions for indication]

1. HER2 positivity testing should be conducted by a pathologist or laboratory with sufficient experience.

2. The efficacy and safety of trastuzumab emtansine in neoadjuvant or adjuvant chemotherapy have not been established.

3. Trastuzumab emtansine should be administered to patients who have received prior treatment with trastuzumab (genetical recombination) and a taxane antineoplastic drug.

[Precautions for dosage and administration]

1. The efficacy and safety of combination therapy with other antineoplastic drugs have not been established.

2. The initial dose should be administered over 90 minutes. If the initial dose is well tolerated, the duration of administration for the second and subsequent doses can be shortened to a minimum of 30 minutes.

3. Treatment interruption, dose reduction, or treatment discontinuation due to an adverse reaction should be decided based on its symptoms, severity, etc., taking into account the following criteria. The dose should not be increased once it is reduced.

Guide for dose reduction

Reduction Step	Dose Level
Starting dose	3.6 mg/kg
First dose reduction	3.0 mg/kg
Second dose reduction	2.4 mg/kg
Requirement for further dose reduction	Discontinue treatment.

(1) Criteria for treatment interruption or treatment discontinuation on the basis of decreased left ventricular ejection fraction (LVEF)

Adverse event		Measures
40% ≤ LVEF ≤ 45%	Absolute decrease is <10% points from baseline.	Continue trastuzumab emtansine treatment. Repeat LVEF assessment within 3 weeks.
	Absolute decrease is ≥10% points from baseline.	Do not administer trastuzumab emtansine treatment. Repeat LVEF assessment within 3 weeks. If LVEF has not recovered to within 10% points from baseline, discontinue trastuzumab emtansine.
LVEF<40%		Do not administer trastuzumab emtansine treatment. Repeat LVEF assessment within 3 weeks. If LVEF is still <40%, discontinue trastuzumab emtansine.
Symptomatic congestive cardiac failure		Discontinue trastuzumab emtansine.

(2) Criteria for treatment interruption, dose reduction, or treatment discontinuation on the basis of increased AST (GOT) or ALT (GPT)

Grade	Measures	
Grade 2 (>3 to 5 × ULN)	Treat at the same dose level.	* Discontinue trastuzumab emtansine if AST (GOT)/ALT (GPT) >3 × ULN and total bilirubin >2 × ULN.
Grade 3 (>5 to 20 × ULN)	Do not administer trastuzumab emtansine until AST (GOT)/ALT (GPT) recovers to Grade ≤2, and then reduce one dose level.	
Grade 4 (>20 × ULN)	Discontinue trastuzumab emtansine.	

(3) Criteria for treatment interruption, dose reduction, or treatment discontinuation on the basis of hyperbilirubinaemia

Grade	Measures	
Grade 2 (>1.5 to 3 × ULN)	Do not administer trastuzumab emtansine until total bilirubin recovers to Grade ≤1, and then treat at the same dose level.	* Discontinue trastuzumab emtansine if AST (GOT)/ALT (GPT) >3 × ULN and total bilirubin >2 × ULN.
Grade 3 (>3 to 10 × ULN)	Do not administer trastuzumab emtansine until total bilirubin recovers to Grade ≤1, and then reduce one dose level.	
Grade 4 (>10 × ULN)	Discontinue trastuzumab emtansine.	

(4) Criteria for treatment interruption or dose reduction on the basis of thrombocytopenia

Grade	Measures
Grade 3 (25,000 to <50,000/mm^3)	Do not administer trastuzumab emtansine until platelet count recovers to Grade ≤1 (≥ 75,000/mm^3), and then treat at the same dose level.
Grade 4 (<25,000/mm^3)	Do not administer trastuzumab emtansine until platelet count recovers to Grade ≤1 (≥ 75,000/mm^3), and then reduce one dose level.

(5) Criteria for treatment interruption on the basis of peripheral neuropathy

Grade	Measures
Grade 3 or 4	Do not administer trastuzumab emtansine until symptoms recover to Grade ≤2, and then treat at the same dose level.

Grades are according to NCI CTCAE (v.4).
ULN: Upper limit of normal

4. Trastuzumab emtansine is reconstituted with the Water for Injection (JP) included in the product package (5 mL for the 100-mg formulation and 8 mL for the 160-mg formulation) to obtain a trastuzumab emtansine (genetical recombination) solution at a concentration of 20 mg/mL. The required volume is then withdrawn from the vial using a syringe, immediately diluted into 250 mL of saline (JP), and intravenously infused.