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APPLICATION NUMBER:

761091Orig1s000

**CLINICAL PHARMACOLOGY AND
BIOPHARMACEUTICS REVIEW(S)**

Office of Clinical Pharmacology
351(k) Biosimilar Review

351(k) BLA Number	761091
Applicant	Celltrion
Resubmission Date	SDN 042: June 15, 2018 SDN 046: September 11, 2018 SDN 053: November 12, 2018
Submission Type	Resubmission
Link to EDR	\\CDSESUB1\evsprod\BLA761091\761091.enx
Brand (Generic) Name	Herzuma (CT-P6)
Dosage Form and Strength	420 mg lyophilized powder in a multiple-dose vial for reconstitution
Route of Administration	Intravenous infusion
Proposed Indication(s)	Adjuvant Breast Cancer • As part of a treatment regimen consisting of doxorubicin, cyclophosphamide, and either paclitaxel or docetaxel • As part of a treatment regimen with docetaxel and carboplatin • As a single agent following multi-modality anthracycline based therapy Metastatic Breast Cancer • In combination with paclitaxel for first-line treatment of HER2-overexpressing metastatic breast cancer • As a single agent for treatment of HER2-overexpressing breast cancer in patients who have received one or more chemotherapy regimens for metastatic disease
Associated IND	119650
Reference Product Information (U.S.-licensed)	
Brand (Generic) Name	Herceptin (Trastuzumab)
Dosage Form and Strength	420 mg of lyophilized drug product in a (b) (4) sterile glass vial
OCP Review Team Signers	
OCP Review Team	Christy S. John, Ph.D. Salaheldin Hamed, Ph.D.
OCP Final Signatory	Nam Atiqur Rahman., Ph.D.

Regulatory History

Celltrion originally submitted the 351(k) BLA 761091 on May 30, 2017. The Office of Clinical Pharmacology recommended approval – refer to the Clinical Pharmacology review of the original BLA submission (DARRTS date: 2/21/2018, Reference ID: 4224199; Appendix). There was an outstanding clinical pharmacology issue related to the long-term stability data supporting sample analysis. A comment was conveyed in the Complete Response Letter as follows:

The applicant has submitted 6-month storage stability data for CT-P6 and US-licensed Herceptin in human serum (Project RFHD2). The 6-month storage stability data did not support the sample analysis using ELISA method (ICD546) for Study CT-P6 1.5, which was around 18-months in duration. Submit to the BLA additional long term storage stability data for the duration of the study covering from the date of sample collection to the last sample analysis date.

Of note, BLA 761091 received a Complete Response Letter on March 30, 2018 due to issues with facilities inspection and product quality. Therefore, Celltrion addressed the issues related to the Agency's Complete Response Letter, including the above outstanding issue, and resubmitted the application on June 15, 2018.

Updated Long-Term Stability Data

In this resubmission (June 15, 2018), the Applicant has submitted long term stability data for Study CT-P5 1.5 at $-80^{\circ}\text{C} \pm 10^{\circ}\text{C}$ and $-25^{\circ}\text{C} \pm 5^{\circ}\text{C}$ for eight months, 12 months, and 15 months using the ELISA method by low quality control (LQC) and high quality control (HQC) samples containing CT-P6 or Herceptin (Validation report, RHDA3).

Reviewer's Comment: The long-term stability data using QC samples stored at $-80^{\circ}\text{C} \pm 10^{\circ}\text{C}$ and $-25^{\circ}\text{C} \pm 5^{\circ}\text{C}$ support storage stability for 434 days (over 14 months). The results showed that all samples met the acceptance criteria (mean CV% $\leq$ (b) (4) %, mean difference from theoretical, DFT% $\pm$ (b) (4) %) recommended by the relevant FDA guidance - *Bioanalytical Method Validation, Guidance for Industry* (FDA, 2018). These stability data over the 14-month duration indicate a high likelihood of successful demonstration of stability for 18 months, which covers the duration from the date of sample collection to the date of last sample analysis. The applicant committed to submit 18-month stability data by February 2019 using ELISA method.

1.1 Recommendations

The Office of Clinical Pharmacology recommends approval of BLA 761091 based on demonstration of PK similarity and no increase in immunogenicity risk between CT-P6 and US-licensed Herceptin.

1.2 Post-Marketing Commitments

Submit to the BLA additional long-term storage stability data for 559 days covering the duration of the study from the date of sample collection to the last sample analysis date.

Signatures

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Appendix: Original BLA 761091 Review

Office of Clinical Pharmacology 351(k) Biosimilar Review

351(k) BLA Number	761091
Applicant	Celltrion
Submission Date	May 30, 2017
Submission Type	Standard
Link to EDR	\\CDSESUB1\evsprod\BLA761091\761091.enx
Brand (Generic) Name	Herzuma (CT-P6)
Dosage Form and Strength	420 mg lyophilized powder in a multiple-dose vial for reconstitution
Route of Administration	Intravenous infusion
Proposed Indication(s)	Adjuvant Breast Cancer • As part of a treatment regimen consisting of doxorubicin, cyclophosphamide, and either paclitaxel or docetaxel • As part of a treatment regimen with docetaxel and carboplatin • As a single agent following multi-modality anthracycline based therapy Metastatic Breast Cancer • In combination with paclitaxel for first-line treatment of HER2-overexpressing metastatic breast cancer • As a single agent for treatment of HER2-overexpressing breast cancer in patients who have received one or more chemotherapy regimens for metastatic disease Metastatic Gastric Cancer • In combination with cisplatin and capecitabine or 5-fluorouracil, for the treatment of patients with HER2 overexpressing metastatic gastric or gastroesophageal junction adenocarcinoma who have not received prior treatment for metastatic disease
Associated IND	119650
Reference Product Information (U.S.-licensed)	
Brand (Generic) Name	Herceptin (Trastuzumab)
Dosage Form and Strength	420 mg of lyophilized drug product in a (b) (4) sterile glass vial
OCP Review Team Signers	
OCP Review Team	Christy S. John, Ph.D. Thiengi M. Thway, Ph.D. Sarah J. Schrieber, Pharm.D.
OCP Final Signatory	Nam Atiqur Rahman., Ph.D.

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1. EXECUTIVE SUMMARY

This Biologic License Application (BLA) for CT-P6 (Herzuma) has been submitted under Section 351(k) of the Public Health Service Act (42 U.S.C. 262(k)). The applicant is seeking approval for CT-P6 as a proposed biosimilar to US-licensed Herceptin licensed under BLA 103792 by Genentech. The applicant is seeking licensure for the treatment of HER2 overexpressing breast cancer, and the treatment of HER2-overexpressing metastatic gastric or gastroesophageal junction adenocarcinoma for which US-licensed Herceptin is currently approved. The applicant submitted pharmacokinetic (PK) similarity data and a comparative clinical study to support a demonstration of no clinically meaningful difference between CT-P6 and US-licensed Herceptin. Study CT-P6 1.5 was a single-dose, randomized, double-blind, 2-arm, parallel group study in 70 healthy male subjects designed to determine the PK similarity of CT-P6, and US-licensed Herceptin following a single 6 mg/kg intravenous dose infused over 90 minutes. The 90% confidence intervals (CI) for all pairwise comparisons of the PK endpoint (AUC_{0-inf}) were within the limits of 80 to 125%. The results of the study established the PK similarity between CT-P6 and US-licensed Herceptin. Overall, Study CT-P6 1.5 supports a demonstration of PK similarity between CT-P6 and US-licensed Herceptin.

The incidence of immunogenicity for CT-P6 and US-licensed Herceptin was compared in a multiple-dose, parallel-arm study in 562 patients with breast cancer (CT-P6 3.2). The results indicate similar incidence and titers of anti-drug antibodies (ADA) for both products. The data indicates that there is no increase in immunogenicity risk for CT-P6 as compared to US-licensed Herceptin.

In conclusion, the PK and immunogenicity results support a demonstration of no clinically meaningful difference between CT-P6 and US-licensed Herceptin and add to the totality of the evidence to support a demonstration of biosimilarity of CT-P6 and US-licensed Herceptin.

1.2 Recommendations

The Office of Clinical Pharmacology recommends approval of CT-P6 based on demonstration of PK similarity and no increase in immunogenicity risk between CT-P6 and US-licensed Herceptin.

Review Issue	Recommendations and Comments
Pivotal evidence of PK similarity	The results of Study CT-P6 1.5 demonstrate PK similarity between CT-P6 and US-licensed Herceptin. The 90% CI of the geometric mean ratio for each product pairwise comparison for the primary PK endpoint of AUC_{0-inf} fell within the margin of 80 to 125%.
Evidence of immunogenicity comparability	The results of Study CT-P6 3.2 indicate similar incidence of ADA for CT-P6 and US-licensed Herceptin.

1.2 Post-Marketing Requirements and Commitments

None.

2. SUMMARY OF CLINICAL PHARMACOLOGY ASSESSMENT

2.1 Clinical Pharmacology and Pharmacokinetics

CT-P6 (Herzuma) is a proposed biosimilar to US-licensed Herceptin. US-licensed Herceptin (trastuzumab) is a humanized IgG₁κ monoclonal antibody directed against an epitope on the extracellular juxtamembrane domain of HER2. Multiple mechanisms of action have been proposed for trastuzumab, including inhibition of HER2 receptor dimerization, increased destruction of the endocytic portion of the HER2 receptor, inhibition of extracellular domain shedding, and activation of cell-mediated immune defenses such as antibody dependent cellular toxicity (ADCC) activity. CT-P6 is produced using a mammalian cell line (b) (4)

. Details on the clinical pharmacology of US-licensed Herceptin can be found in the product label (USPI).

In Study CT-P6 1.5, the 90% CI for the geometric mean ratios of the primary PK endpoint of AUC_{0-inf} and secondary PK endpoint of C_{max} were within the limits of 80% to 125% as summarized in **Table 1**.

Table 1. Statistical analysis for pharmacokinetic similarity (Study CT-P6 1.5)

Comparison	Geometric Mean Ratio* (90% CI)	
	AUC_{0-inf}	C_{max}
CT-P6 vs US-licensed Herceptin	94.4 (88.2-101.0)	96.7 (91.1-102.7)

*expressed as a percent

Overall, the submitted clinical pharmacology study adequately demonstrated similarity of PK between CT-P6 and US-licensed Herceptin. The PK results support a demonstration of no clinically meaningful differences between CT-P6 and US-licensed Herceptin, and add to the totality of the evidence to support a demonstration of biosimilarity of CT-P6 and US-licensed Herceptin.

The incidence of immunogenicity for CT-P6 and US-licensed Herceptin was compared in a multiple-dose, parallel-arm study in 562 patients with metastatic breast cancer (CT-P6 3.2). The results indicate similar incidence and titers of ADAs for both products. The data indicates that there is no increase in immunogenicity risk for CT-P6 as compared to US-licensed Herceptin, and supports the demonstration of no clinically meaningful differences between CT-P6 and US-licensed Herceptin.

2.2 Outstanding Issues

The following clinical pharmacology comment is to be included in the action letter:

The applicant has submitted 6-month storage stability data for CT-P6 and US-licensed Herceptin in human serum (Project RFHD2). The 6-month storage stability data did not support the sample analysis using ELISA method (ICD546) for Study CT-P6 1.5, which was around 18-months in duration. Submit to the BLA additional long term storage stability data for the duration of the study covering from the date of sample collection to the last sample analysis date.

3. COMPREHENSIVE CLINICAL PHARMACOLOGY REVIEW

3.1 Regulatory Background

3.1.1 Describe relevant regulatory history for the review of this 351(k) BLA.

CT-P6 is a proposed biosimilar to US-licensed Herceptin. The applicant is seeking licensure for the treatment of adjuvant or metastatic breast cancer and metastatic gastric or gastroesophageal adenocarcinoma indications for which US-licensed Herceptin is currently approved.

3.2 Clinical Pharmacology Review Questions

3.2.1 What are the design features of the clinical pharmacology and/or clinical studies to support biosimilarity?

The applicant conducted a PK similarity Study CT-P6 1.5 and one comparative clinical study, as is described in **Table 2**.

Table 2. Summary of relevant clinical studies

Protocol #	Title	Subjects	Objectives	Route/Dose/Duration
PK Similarity Study				
CT-P6 1.5 (PK Similarity Study)	Phase 1, randomized (1:1), controlled, 2-arm, parallel group, double blind, single dose study to compare PK and immunogenicity of CT-P6 vs US-licensed Herceptin	Healthy male (N=70)	PK similarity	IV, Single dose of 6 mg/kg IV infusion for 90 min
Comparative Clinical Study				
CT-P6 3.2 (Clinical efficacy and safety study)	A phase 3, double-blind, randomized, parallel-group, active-controlled study to compare the efficacy and safety of CT-P6 and US-licensed Herceptin as neoadjuvant and adjuvant treatment in patients with HER2-positive early breast cancer	Early breast cancer female, (N=562)	Efficacy, safety, immunogenicity	IV infusion 8 mg/kg body weight on Day 1 of Neoadjuvant Period Cycle 1, 6 mg/kg body weight repeated every 3 weeks (Neoadjuvant Period and Adjuvant Period) IV infusion

PK: pharmacokinetics; IV, intravenous

Study CT-P6 1.5 was used to support PK similarity. This study was a single-dose, randomized, double-blind, 2-arm, parallel-group study designed to compare the pharmacokinetic profiles of CT-P6 (n = 35) and US-licensed Herceptin (n = 35), administered as a single 6 mg/kg

intravenous dose infused over 90 minutes to healthy male volunteers. PK similarity was concluded if the 90% CI of the geometric mean ratio for AUC_{0-inf} between CT-P6 and US-licensed Herceptin were within the limits of 80% to 125%.

The study design of Study CT-P6 1.5 is considered adequate due to the following reasons:

1. A single-dose, parallel group design is appropriate for trastuzumab because the product has a long half-life (ranging from 2 to 12 days).
2. A study in healthy subjects is considered safe and more sensitive compared with that in patients with potentially confounding factors such as underlying disease, concomitant medications, and other factors.
3. Considering PK assay sensitivity, dose-exposure linearity, and tolerability, a single IV dose of 6 mg/kg trastuzumab is considered appropriate for a PK similarity study.

CT-P6 3.2 was a comparative clinical study in HER2-positive early breast cancer patients. Refer to Clinical review for further details.

3.2.2 What are the endpoints in the clinical pharmacology and/or clinical studies to support biosimilarity?

In Study CT-P6 1.5, the PK similarity criteria for AUC_{0-inf} , and C_{max} were that the 90% CI of the geometric mean ratio should lie within 80-125%. This margin proposed by the applicant was acceptable.

- PK serum samples were collected on Day 1 at predose 0.75, 1.5, 3, 6, 12, 24, 48, 96, 168, 336, 504, 672, 1176, and 1680 hours post dose.

3.2.3 Are the pharmacologically active moieties of the proposed biosimilar and the reference product in plasma (or other biological matrix) appropriately identified and measured to assess the PK parameters?

Yes. Trastuzumab concentrations were measured in serum by a validated enzyme-linked immunosorbent assay (ELISA). See **Section 4.1.1** for details.

3.2.4 Is PK similarity met?

Yes, PK similarity between CT-P6 and US-licensed Herceptin was demonstrated where the 90% CI for the geometric mean ratios of the primary PK endpoint of AUC_{0-inf} and secondary PK endpoint of C_{max} were within the limits of 80% to 125% (**Table 1**). Also, as shown in **Figure 1**, the mean trastuzumab concentration vs time profiles of CT-P6 and US-licensed Herceptin overlay each other. A summary of the 6 mg/kg single IV dose PK parameters from Study CT-P6 1.5 for each product is shown in **Table 3**.

Figure 1. Mean serum trastuzumab concentration vs. time profile (Study CT-P6 1.5)

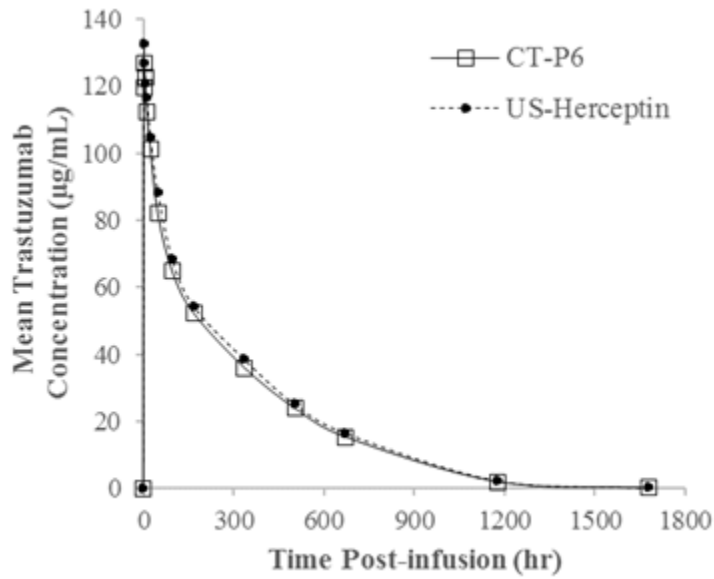


Table 3. Summary of PK parameters (Study CT-P6 1.5)

PK parameters	Geometric Mean (% CV)	
	CT-P6 (n=35)	US-licensed Herceptin (n=35)
C_{max} (µg/mL)	131 (14)	135 (16)
AUC_{0-t} (µg /mL*hr)	32634 (16)	34482 (18)
AUC_{0-inf} (µg/mL*hr)	32863 (16)	34821 (18)
$t_{1/2}$ (hr)	167 (21)	175 (16)

% CV: coefficient of variation, $t_{1/2}$ = elimination half-life,

Immunogenicity

3.2.6 Is the immunogenicity assay capable of detecting the anti-drug antibodies (ADA) in the presence of concentration of product in the study samples?

Yes. For study CT-P6 3.2, detection, confirmation, and titration of ADAs were determined utilizing a Meso Scale Discovery method (Method # ICDIM 192) at (b) (4). The method sensitivity, low positive control, and high positive control of the ADA method were 0.433 ng/mL, 3.51 ng/mL, and 2000 ng/mL, respectively. The drug tolerance limits of the ADA method were 2 or 25 µg/mL respectively when tested with positive control concentrations of 3.51 or 100 ng/mL. Serum trastuzumab concentrations are expected to be < 2.3 µg/mL on 49 days post IV infusion indicating that ADA method is drug-tolerant for the sampling schedules listed in **Section 3.2.7**.

Refer to the immunogenicity assay review by the Office of Biological Products for details regarding the method.

3.2.7 Is the sampling plan adequate to capture baseline, early onset, and dynamic profile (transient or persistent) of anti-drug antibodies (ADA) formation?

The sampling schedules in the studies were appropriate to minimize interference from the presence of the product in the samples, if the ADA method is not drug-tolerant. The sampling schedules for the studies were as follows:

- Study CT-P6 1.5: Pre-dose on Day 1 and post-dose on Days 50 and 71
- Study CT-P6 3.2:
 - Neoadjuvant Period: Pre-dose (baseline), Cycle 4, and End of treatment
 - Adjuvant Period: Pre-dose on Day 1 of Cycle 1, after Cycle 3, after Cycle 6, and End of treatment.
 - Follow-up Period: Every 3 months up to 1 year

3.2.8 What is the incidence of anti-drug antibodies (ADA)? (Provide the incidence of pre-existing antibodies at baseline and the incidence of ADA throughout the study.)

In Study CT-P6 1.5, none of the subjects had a positive ADA test result at the time points assessed.

In Study CT-P6 3.2, a low incidence of ADA, which were not neutralizing, was observed at pre-dose (baseline) for patients randomized to CT-P6 and US-licensed Herceptin (**Table 4**). No treatment-induced ADA were observed in the study (**Table 4**).

Table 4. Immunogenicity results from Study CT-P6 3.2

Product	N	Baseline ADA*	Treatment- induced ADA
CT-P6	271	1.5% (4/271)	0%
US-licensed Herceptin	278	2.9% (8/278)	0%

*No samples were positive for neutralizing ADAs

The data indicate that there is no increase in immunogenicity risk for CT-P6 as compared to US-licensed Herceptin, and supports the demonstration that there are no clinically meaningful differences between CT-P6 and US-licensed Herceptin.

3.2.9 Do the anti-drug antibodies (ADA) have neutralizing activity?

No patients tested positive for neutralizing antibodies (**Table 4**).

3.2.10 What is the impact of anti-drug antibodies (ADA) on the PK, activity, and safety of the therapeutic protein?

No apparent impact of ADA on PK, activity, or safety endpoints was observed from Study CT-P6 3.2.

4. APPENDICES

4.1 Summary of Bioanalytical Method Validation and Performance

4.1.1 Pharmacokinetics

4.1.1.1 How are the concentrations of the pharmacologically active moieties (parent and/or any relevant catabolites) measured in the plasma and other matrices in the clinical pharmacology studies?

For the PK similarity study (CT-P6 1.5), serum trastuzumab concentrations measured using a validated ELISA method (ICD 546) were suitable for assessment of PK similarity. Both the method validation entitled “An ELISA Method for the Quantitation of CT-P6 in Human Serum” and sample analysis for the study were performed at (b) (4). In this method, histidine-tagged human HER2 (b) (4) coated in 96-well plate was used to capture serum CT-P6 or US-licensed Herceptin and a HRP-conjugated goat anti-human IgG Fc antibody (b) (4) was used to detect the bound analyte. Table 5 shows the summary of ELISA method performance in quantification of serum CT-P6 and US-licensed Herceptin during the method validation.

Table 5. Summary of method validation of an ELISA to measure CT-P6 and US-licensed Herceptin in human serum

(b) (4) Project Codes	RHDA7, RHDA 8, and RHDA10	
Method ID	ICD 546 (Versions 3.01 - 3.03)	
Material for calibration curve & concentration	CT-P6, Lot 13A3C005 at 21.0 mg/mL after reconstitution US-licensed Herceptin, Lot 626323, 21.0 mg/mL after reconstitution	
Minimum required dilution (MRD)	1:100 in buffer (Blocker™ Casein in PBS)	
Matrix	Human serum	
Source & lot of reagents	Histidine-tagged human HER2 (b) (4) Lot: C49p1-68GS1-B HRP-conjugated goat anti-human IgG Fc antibody (b) (4) (b) (4), Lot:131321	
Sample Volume (µL)	20 µL	
Sample Storage	-80 °C ± 10 °C	
Validated assay range (LLOQ to ULOQ)	200 (LLOQ) - 6400 (ULOQ) ng/mL	
Regression, Weighting	four-parameter logistic, 1/Y ²	
QC Concentrations	200, 400, 600, 1500, 4800, 5500, and 6400 ng/mL 1050 mcg/ml as dilution QC diluted at 1/1000	
Method validation summary		
Standard curve performance during accuracy & precision	Number of standard calibrators from LLOQ to ULOQ: 6	Yes
	Cumulative accuracy (%bias) from LLOQ to ULOQ: -6.4 to 5.6%	Yes
	Cumulative precision (%CV) from LLOQ to ULOQ: ≤5%	Yes

QCs performance during accuracy & precision	Cumulative accuracy (%bias) in 7 QCs* CT-P6 QCs: -2.2 to 12.6% US-licensed Herceptin QCs: -3.7 to 8.0%	Yes
	Inter-assay %CV in 7 QCs: CT-P6 QCs: ≤ 18% US-licensed Herceptin QCs: ≤ 17%	Yes
	Percent total error (TE) in 7 QCs: CT-P6 QCs: ≤ 28% US-licensed Herceptin QCs: ≤ 23%	Yes
Selectivity	Tested in these serum types at blank (0 ng/mL), LLOQ and HQC levels: Unfortified healthy human serum (HHS) – 10 lots • HSS fortified with CT-P6 • HSS fortified with US-Herceptin Unfortified breast cancer serum (BCS) – 10 lots • BCS fortified with CT-P6 • BCS fortified with US-Herceptin All lots meet the acceptance criteria.	Yes
Dilutional Linearity & hook effect	Linear within 500- to 2000- fold dilutions. Tested with 1050 mcg/mL of CT-P6 or US-licensed Herceptin No hook effect	Yes
Hemolysis	No effect from hemolysis on the quantitation of CT-P6 or US-licensed Herceptin	Yes
Lipemia	No effect from lipemia on the quantitation of CT-P6 or US-licensed Herceptin	Yes
Interference	No interference from sHER2 when tested up to 1000 ng/mL of Her2 with 600 ng/mL of CT-P6 or US-licensed Herceptin	Yes, as expected
Bench-top/process stability	CT-P6: 26 hours at room temperature in serum, US-licensed Herceptin: 24 hours at room temperature in serum	Yes
Freeze-thaw stability	Up to 10 cycles thawed at room temperature for both CT-P6 and US-licensed Herceptin	Yes
Frozen serum storage stability (days)	CT-P6: 35 days at -80 °C ± 10 °C and -25 °C ± 5 °C US-licensed Herceptin: 17 days at -80 °C ± 10 °C and -25 °C ± 5 °C	No**

*One additional QC each near LLOQ and ULOQ.

Long-term stability data has not been provided refer to **Table 6.

Performance of the validated ELISA method (ICD 546) during the sample analysis for study CT-P6 1.5 is summarized in **Table 6**.

Table 6. Summary of ELISA method performance in support of study CT-P6 1.5

(b) (4) Project Code	RFHD2	Acceptability
Assay passing rate	• 87% of the assay met the method acceptance criteria.	Yes
Standard curve performance	• Cumulative bias range: -7.07 to 7.13% • Cumulative precision: ≤ 5%CV	Yes
QC performance	• Cumulative bias range: -0.7 to 8.6% • Cumulative precision: ≤ 13%CV • TE: ≤ 20%	Yes

Study sample analysis/ stability	Refer to Section 2.2 for the duration to establish serum storage stability.

Initially, serum trastuzumab concentrations from study CT-P6 1.5 were determined using a validated GyrosLab® -based immunoassay at (b) (4). This method (M08.CT-P6 or Trastuzumab.huse.1) was developed at (b) (4) and transferred to (b) (4) where the validated range was further extended. Upon review of the submission, this method was found not suitable to accurately measure serum trastuzumab concentrations; see **Table 7** below for a summary of this method. In GyrosLab®-based immunoassay, biotinylated- antibody specific to the cytoplasmic domain of HER-2 were first allowed to pass through the microstructures containing streptavidin-coated beads in Bioaffy 1000 CDs, followed by full-length HER-2. Then, the serum samples containing trastuzumab were allowed to pass through these microstructures where trastuzumab then bound to HER-2. Immobilized trastuzumab was detected by Alexa Fluor® 647-conjugated purified mouse anti-human IgG1 antibody. Wash steps were performed between each reagent/ sample addition to remove any unbound material. The CD was read by a laser that excited the Alexa Fluor® 647, producing a fluorescent signal that was directly proportional to the concentration of trastuzumab. **Table 7** shows brief summary review explaining the deficiency of the method resulted in under-quantification of serum trastuzumab.

Table 7. Brief summary of GyrosLab® -based immunoassay method used to support study CT-P6 1.5

Bioanalytical method review summary	GyrosLab® -based immunoassay method had a deficiency since serum trastuzumab concentrations were not accurately measured within the validated calibration range due to following 2 main reasons; 1) the amount of capture reagents allowed to pass through the microstructures were limited compared to trastuzumab concentrations even after MRD, and 2) the presence of additional capture step (Capture 1 to capture 2) might require longer incubation time to reach an optimal equilibrium than GyrosLab® allowed.
(b) (4) Project code	RBNU2
(b) (4) method #	ICD 546
Material for calibration curve & concentration	CT-P6, Lot 13A3C005 at 21.0 mg/mL US-licensed Herceptin, Lot 554763, 21.0 mg/mL
Validated assay range	5.00 (LLOQ) – 750 (ULOQ) mcg/mL
Minimum required dilution (MRD)	1:1000 in sample diluent (filtered Superblock with 250 mcg/mL mouse IgG)
Source & lot of reagents	- Capture 1: Biotinylated- c-erbB-2/Her-2/neu Ab-17, clone e2-4001+3B5 (b) (4), Lot: NB#9145-67-01 - Capture 2: Recombinant Her-2 (b) (4) Lot: 111313 - Detection: Alexa Fluor® 647-conjugated purified mouse anti-human IgG (b) (4), Lot: NB#9145-84-01
Regression model & weighting	5-Parameter logistic, weighting of 1/Y ²

As result of the inadequacies of the GyrosLab®-based immunoassay, 1) additional details regarding method validation are not included and 2) serum trastuzumab concentrations determined using a validated ELISA method described in **Table 5** were reviewed and used for the PK similarity assessment.

This is a representation of an electronic record that was signed electronically. Following this are manifestations of any and all electronic signatures for this electronic record.

/s/

CHRISTY S JOHN
11/28/2018

SALAHELDIN HAMED
11/28/2018

NAM ATIQR RAHMAN
11/29/2018

Office of Clinical Pharmacology
351(k) Biosimilar Review

351(k) BLA Number	761091
Applicant	Celltrion
Submission Date	May 30, 2017
Submission Type	Standard
Link to EDR	\\CDSESUB1\evsprod\BLA761091\761091.enx
Brand (Generic) Name	Herzuma (CT-P6)
Dosage Form and Strength	420 mg lyophilized powder in a multiple-dose vial for reconstitution
Route of Administration	Intravenous infusion
Proposed Indication(s)	Adjuvant Breast Cancer • As part of a treatment regimen consisting of doxorubicin, cyclophosphamide, and either paclitaxel or docetaxel • As part of a treatment regimen with docetaxel and carboplatin • As a single agent following multi-modality anthracycline based therapy Metastatic Breast Cancer • In combination with paclitaxel for first-line treatment of HER2-overexpressing metastatic breast cancer • As a single agent for treatment of HER2-overexpressing breast cancer in patients who have received one or more chemotherapy regimens for metastatic disease Metastatic Gastric Cancer • In combination with cisplatin and capecitabine or 5-fluorouracil, for the treatment of patients with HER2 overexpressing metastatic gastric or gastroesophageal junction adenocarcinoma who have not received prior treatment for metastatic disease
Associated IND	119650
Reference Product Information (U.S.-licensed)	
Brand (Generic) Name	Herceptin (Trastuzumab)
Dosage Form and Strength	420 mg of lyophilized drug product in a (b) (4) sterile glass vial
OCP Review Team Signers	
OCP Review Team	Christy S. John, Ph.D. Thiengi M. Thway, Ph.D. Sarah J. Schrieber, Pharm.D.
OCP Final Signatory	Nam Atiqur Rahman., Ph.D.

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1. EXECUTIVE SUMMARY

This Biologic License Application (BLA) for CT-P6 (Herzuma) has been submitted under Section 351(k) of the Public Health Service Act (42 U.S.C. 262(k)). The applicant is seeking approval for CT-P6 as a proposed biosimilar to US-licensed Herceptin licensed under BLA 103792 by Genentech. The applicant is seeking licensure for the treatment of HER2 overexpressing breast cancer, and the treatment of HER2-overexpressing metastatic gastric or gastroesophageal junction adenocarcinoma for which US-licensed Herceptin is currently approved. The applicant submitted pharmacokinetic (PK) similarity data and a comparative clinical study to support a demonstration of no clinically meaningful difference between CT-P6 and US-licensed Herceptin. Study CT-P6 1.5 was a single-dose, randomized, double-blind, 2-arm, parallel group study in 70 healthy male subjects designed to determine the PK similarity of CT-P6, and US-licensed Herceptin following a single 6 mg/kg intravenous dose infused over 90 minutes. The 90% confidence intervals (CI) for all pairwise comparisons of the PK endpoint (AUC_{0-inf}) were within the limits of 80 to 125%. The results of the study established the PK similarity between CT-P6 and US-licensed Herceptin. Overall, Study CT-P6 1.5 supports a demonstration of PK similarity between CT-P6 and US-licensed Herceptin.

The incidence of immunogenicity for CT-P6 and US-licensed Herceptin was compared in a multiple-dose, parallel-arm study in 562 patients with breast cancer (CT-P6 3.2). The results indicate similar incidence and titers of anti-drug antibodies (ADA) for both products. The data indicates that there is no increase in immunogenicity risk for CT-P6 as compared to US-licensed Herceptin.

In conclusion, the PK and immunogenicity results support a demonstration of no clinically meaningful difference between CT-P6 and US-licensed Herceptin and add to the totality of the evidence to support a demonstration of biosimilarity of CT-P6 and US-licensed Herceptin.

1.1 Recommendations

The Office of Clinical Pharmacology recommends approval of CT-P6 based on demonstration of PK similarity and no increase in immunogenicity risk between CT-P6 and US-licensed Herceptin.

Review Issue	Recommendations and Comments
Pivotal evidence of PK similarity	The results of Study CT-P6 1.5 demonstrate PK similarity between CT-P6 and US-licensed Herceptin. The 90% CI of the geometric mean ratio for each product pairwise comparison for the primary PK endpoint of AUC _{0-inf} fell within the margin of 80 to 125%.
Evidence of immunogenicity comparability	The results of Study CT-P6 3.2 indicate similar incidence of ADA for CT-P6 and US-licensed Herceptin.

1.2 Post-Marketing Requirements and Commitments

None.

2. SUMMARY OF CLINICAL PHARMACOLOGY ASSESSMENT

2.1 Clinical Pharmacology and Pharmacokinetics

CT-P6 (Herzuma) is a proposed biosimilar to US-licensed Herceptin. US-licensed Herceptin (trastuzumab) is a humanized IgG₁κ monoclonal antibody directed against an epitope on the extracellular juxtamembrane domain of HER2. Multiple mechanisms of action have been proposed for trastuzumab, including inhibition of HER2 receptor dimerization, increased destruction of the endocytic portion of the HER2 receptor, inhibition of extracellular domain shedding, and activation of cell-mediated immune defenses such as antibody dependent cellular toxicity (ADCC) activity. CT-P6 is produced using a mammalian cell line (b) (4)

. Details on the clinical pharmacology of US-licensed Herceptin can be found in the product label (USPI).

In Study CT-P6 1.5, the 90% CI for the geometric mean ratios of the primary PK endpoint of AUC_{0-inf} and secondary PK endpoint of C_{max} were within the limits of 80% to 125% as summarized in **Table 1**.

Table 1. Statistical analysis for pharmacokinetic similarity (Study CT-P6 1.5)

Comparison	Geometric Mean Ratio* (90% CI)	
	AUC _{0-inf}	C _{max}
CT-P6 vs US-licensed Herceptin	94.4 (88.2-101.0)	96.7 (91.1-102.7)

*expressed as a percent

Overall, the submitted clinical pharmacology study adequately demonstrated similarity of PK between CT-P6 and US-licensed Herceptin. The PK results support a demonstration of no clinically meaningful differences between CT-P6 and US-licensed Herceptin, and add to the totality of the evidence to support a demonstration of biosimilarity of CT-P6 and US-licensed Herceptin.

The incidence of immunogenicity for CT-P6 and US-licensed Herceptin was compared in a multiple-dose, parallel-arm study in 562 patients with metastatic breast cancer (CT-P6 3.2). The results indicate similar incidence and titers of ADAs for both products. The data indicates that there is no increase in immunogenicity risk for CT-P6 as compared to US-licensed Herceptin, and supports the demonstration of no clinically meaningful differences between CT-P6 and US-licensed Herceptin.

2.2 Outstanding Issues

The following clinical pharmacology comment is to be included in the action letter:

The applicant has submitted 6-month storage stability data for CT-P6 and US-licensed Herceptin in human serum (Project RFHD2). The 6-month storage stability data did not support the sample analysis using ELISA method (ICD546) for Study CT-P6 1.5, which was around 18-months in duration. Submit to the BLA additional long term storage stability data for the duration of the study covering from the date of sample collection to the last sample analysis date.

3. COMPREHENSIVE CLINICAL PHARMACOLOGY REVIEW

3.1 Regulatory Background

3.1.1 Describe relevant regulatory history for the review of this 351(k) BLA.

CT-P6 is a proposed biosimilar to US-licensed Herceptin. The applicant is seeking licensure for the treatment of adjuvant or metastatic breast cancer and metastatic gastric or gastroesophageal adenocarcinoma indications for which US-licensed Herceptin is currently approved.

3.2 Clinical Pharmacology Review Questions

3.2.1 What are the design features of the clinical pharmacology and/or clinical studies to support biosimilarity?

The applicant conducted a PK similarity Study CT-P6 1.5 and one comparative clinical study, as is described in **Table 2**.

Table 2. Summary of relevant clinical studies

Protocol #	Title	Subjects	Objectives	Route/Dose/Duration
PK Similarity Study				
CT-P6 1.5 (PK Similarity Study)	Phase 1, randomized (1:1), controlled, 2-arm, parallel group, double blind, single dose study to compare PK and immunogenicity of CT-P6 vs US-licensed Herceptin	Healthy male (N=70)	PK similarity	IV, Single dose of 6 mg/kg IV infusion for 90 min
Comparative Clinical Study				
CT-P6 3.2 (Clinical efficacy and safety study)	A phase 3, double-blind, randomized, parallel-group, active-controlled study to compare the efficacy and safety of CT-P6 and US-licensed Herceptin as neoadjuvant and adjuvant treatment in patients with HER2-positive early breast cancer	Early breast cancer female, (N=562)	Efficacy, safety, immunogenicity	IV infusion 8 mg/kg body weight on Day 1 of Neoadjuvant Period Cycle 1, 6 mg/kg body weight repeated every 3 weeks (Neoadjuvant Period and Adjuvant Period) IV infusion

PK: pharmacokinetics; IV, intravenous

Study CT-P6 1.5 was used to support PK similarity. This study was a single-dose, randomized, double-blind, 2-arm, parallel-group study designed to compare the pharmacokinetic profiles of CT-P6 (n = 35) and US-licensed Herceptin (n = 35), administered as a single 6 mg/kg intravenous dose infused over 90 minutes to healthy male volunteers. PK similarity was concluded if the 90% CI of the geometric mean ratio for AUC_{0-inf} between CT-P6 and US-licensed Herceptin were within the limits of 80% to 125%.

The study design of Study CT-P6 1.5 is considered adequate due to the following reasons:

1. A single-dose, parallel group design is appropriate for trastuzumab because the product has a long half-life (ranging from 2 to 12 days).
2. A study in healthy subjects is considered safe and more sensitive compared with that in patients with potentially confounding factors such as underlying disease, concomitant medications, and other factors.
3. Considering PK assay sensitivity, dose-exposure linearity, and tolerability, a single IV dose of 6 mg/kg trastuzumab is considered appropriate for a PK similarity study.

CT-P6 3.2 was a comparative clinical study in HER2-positive early breast cancer patients. Refer to Clinical review for further details.

3.2.2 What are the endpoints in the clinical pharmacology and/or clinical studies to support biosimilarity?

In Study CT-P6 1.5, the PK similarity criteria for $AUC_{0-\text{inf}}$ and C_{max} were that the 90% CI of the geometric mean ratio should lie within 80-125%. This margin proposed by the applicant was acceptable.

- PK serum samples were collected on Day 1 at predose 0.75, 1.5, 3, 6, 12, 24, 48, 96, 168, 336, 504, 672, 1176, and 1680 hours post dose.

3.2.3 Are the pharmacologically active moieties of the proposed biosimilar and the reference product in plasma (or other biological matrix) appropriately identified and measured to assess the PK parameters?

Yes. Trastuzumab concentrations were measured in serum by a validated enzyme-linked immunosorbent assay (ELISA). See **Section 4.1.1** for details.

3.2.4 Is PK similarity met?

Yes, PK similarity between CT-P6 and US-licensed Herceptin was demonstrated where the 90% CI for the geometric mean ratios of the primary PK endpoint of $AUC_{0-\text{inf}}$ and secondary PK endpoint of C_{max} were within the limits of 80% to 125% (**Table 1**). Also, as shown in **Figure 1**, the mean trastuzumab concentration vs time profiles of CT-P6 and US-licensed Herceptin overlay each other. A summary of the 6 mg/kg single IV dose PK parameters from Study CT-P6 1.5 for each product is shown in **Table 3**.

Figure 1. Mean serum trastuzumab concentration vs. time profile (Study CT-P6 1.5)

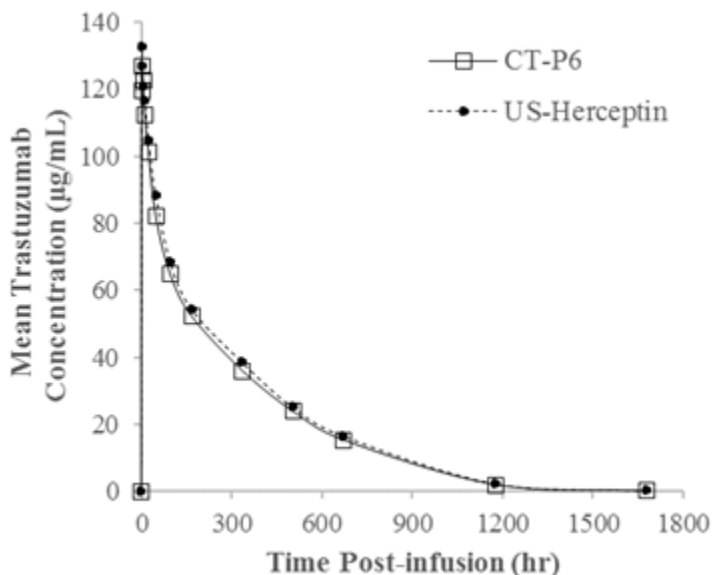


Table 3. Summary of PK parameters (Study CT-P6 1.5)

PK parameters	Geometric Mean (% CV)	
	CT-P6 (n=35)	US-licensed Herceptin (n=35)
C _{max} (µg/mL)	131 (14)	135 (16)
AUC _{0-t} (µg /mL*hr)	32634 (16)	34482 (18)
AUC _{0-inf} (µg/mL*hr)	32863 (16)	34821 (18)
t _{1/2} (hr)	167 (21)	175 (16)

% CV: coefficient of variation, t_{1/2} = elimination half-life,

Immunogenicity

3.2.6 Is the immunogenicity assay capable of detecting the anti-drug antibodies (ADA) in the presence of concentration of product in the study samples?

Yes. For study CT-P6 3.2, detection, confirmation, and titration of ADAs were determined utilizing a Meso Scale Discovery method (Method # ICDIM 192) at (b) (4)

The method sensitivity, low positive control, and high positive control of the ADA method were 0.433 ng/mL, 3.51 ng/mL, and 2000 ng/mL, respectively. The drug tolerance limits of the ADA method were 2 or 25 µg/mL respectively when tested with positive control concentrations of 3.51 or 100 ng/mL. Serum trastuzumab concentrations are expected to be < 2.3 µg/mL on 49 days post IV infusion indicating that ADA method is drug-tolerant for the sampling schedules listed in Section 3.2.7.

Refer to the immunogenicity assay review by the Office of Biological Products for details regarding the method.

3.2.7 Is the sampling plan adequate to capture baseline, early onset, and dynamic profile (transient or persistent) of anti-drug antibodies (ADA) formation?

The sampling schedules in the studies were appropriate to minimize interference from the presence of the product in the samples, if the ADA method is not drug-tolerant. The sampling schedules for the studies were as follows:

- Study CT-P6 1.5: Pre-dose on Day 1 and post-dose on Days 50 and 71
- Study CT-P6 3.2:
 - Neoadjuvant Period: Pre-dose (baseline), Cycle 4, and End of treatment
 - Adjuvant Period: Pre-dose on Day 1 of Cycle 1, after Cycle 3, after Cycle 6, and End of treatment.
 - Follow-up Period: Every 3 months up to 1 year

3.2.8 What is the incidence of anti-drug antibodies (ADA)? (Provide the incidence of pre-existing antibodies at baseline and the incidence of ADA throughout the study.)

In Study CT-P6 1.5, none of the subjects had a positive ADA test result at the time points assessed.

In Study CT-P6 3.2, a low incidence of ADA, which were not neutralizing, was observed at pre-dose (baseline) for patients randomized to CT-P6 and US-licensed Herceptin (**Table 4**). No treatment-induced ADA were observed in the study (**Table 4**).

Table 4. Immunogenicity results from Study CT-P6 3.2

Product	N	Baseline ADA*	Treatment-induced ADA
CT-P6	271	1.5% (4/271)	0%
US-licensed Herceptin	278	2.9% (8/278)	0%

*No samples were positive for neutralizing ADAs

The data indicate that there is no increase in immunogenicity risk for CT-P6 as compared to US-licensed Herceptin, and supports the demonstration that there are no clinically meaningful differences between CT-P6 and US-licensed Herceptin.

3.2.9 Do the anti-drug antibodies (ADA) have neutralizing activity?

No patients tested positive for neutralizing antibodies (**Table 4**).

3.2.10 What is the impact of anti-drug antibodies (ADA) on the PK, activity, and safety of the therapeutic protein?

No apparent impact of ADA on PK, activity, or safety endpoints was observed from Study CT-P6 3.2.

4. APPENDICES

4.1 Summary of Bioanalytical Method Validation and Performance

4.1.1 Pharmacokinetics

4.1.1.1 How are the concentrations of the pharmacologically active moieties (parent and/or any relevant catabolites) measured in the plasma and other matrices in the clinical pharmacology studies?

For the PK similarity study (CT-P6 1.5), serum trastuzumab concentrations measured using a validated ELISA method (ICD 546) were suitable for assessment of PK similarity. Both the method validation entitled “An ELISA Method for the Quantitation of CT-P6 in Human Serum” and sample analysis for the study were performed at (b) (4). In this method, histidine-tagged human HER2 (b) (4) coated in 96-well plate was used to capture serum CT-P6 or US-licensed Herceptin and a HRP-conjugated goat anti-human IgG Fc antibody (b) (4) was used to detect the bound analyte. Table 5 shows the summary of ELISA method performance in quantification of serum CT-P6 and US-licensed Herceptin during the method validation.

Table 5. Summary of method validation of an ELISA to measure CT-P6 and US-licensed Herceptin in human serum

(b) (4) Project Codes	RHDA7, RHDA 8, and RHDA10	
Method ID	ICD 546 (Versions 3.01 - 3.03)	
Material for calibration curve & concentration	CT-P6, Lot 13A3C005 at 21.0 mg/mL after reconstitution US-licensed Herceptin, Lot 626323, 21.0 mg/mL after reconstitution	
Minimum required dilution (MRD)	1:100 in buffer (Blocker™ Casein in PBS)	
Matrix	Human serum	
Source & lot of reagents	Histidine-tagged human HER2 (b) (4), Lot: C49p1-68GS1-B HRP-conjugated goat anti-human IgG Fc antibody (b) (4) (b) (4), Lot:131321	
Sample Volume (µL)	20 µL	
Sample Storage	-80 °C ± 10 °C	
Validated assay range (LLOQ to ULOQ)	200 (LLOQ) - 6400 (ULOQ) ng/mL	
Regression, Weighting	four-parameter logistic, 1/Y ²	
QC Concentrations	200, 400, 600, 1500, 4800, 5500, and 6400 ng/mL 1050 mcg/ml as dilution QC diluted at 1/1000	
Method validation summary		
Standard curve performance during accuracy & precision	Number of standard calibrators from LLOQ to ULOQ: 6	Yes
	Cumulative accuracy (%bias) from LLOQ to ULOQ: -6.4 to 5.6%	Yes
	Cumulative precision (%CV) from LLOQ to ULOQ: ≤5%	Yes

QCs performance during accuracy & precision	Cumulative accuracy (%bias) in 7 QCs* CT-P6 QCs: -2.2 to 12.6% US-licensed Herceptin QCs: -3.7 to 8.0%	Yes
	Inter-assay %CV in 7 QCs: CT-P6 QCs: ≤ 18% US-licensed Herceptin QCs: ≤ 17%	Yes
	Percent total error (TE) in 7 QCs: CT-P6 QCs: ≤ 28% US-licensed Herceptin QCs: ≤ 23%	Yes
Selectivity	Tested in these serum types at blank (0 ng/mL), LLOQ and HQC levels: Unfortified healthy human serum (HHS) – 10 lots • HSS fortified with CT-P6 • HSS fortified with US-Herceptin Unfortified breast cancer serum (BCS) – 10 lots • BCS fortified with CT-P6 • BCS fortified with US-Herceptin All lots meet the acceptance criteria.	Yes
Dilutional Linearity & hook effect	Linear within 500- to 2000- fold dilutions. Tested with 1050 mcg/mL of CT-P6 or US-licensed Herceptin No hook effect	Yes
Hemolysis	No effect from hemolysis on the quantitation of CT-P6 or US-licensed Herceptin	Yes
Lipemia	No effect from lipemia on the quantitation of CT-P6 or US-licensed Herceptin	Yes
Interference	No interference from sHER2 when tested up to 1000 ng/mL of Her2 with 600 ng/mL of CT-P6 or US-licensed Herceptin	Yes, as expected
Bench-top/process stability	CT-P6: 26 hours at room temperature in serum, US-licensed Herceptin: 24 hours at room temperature in serum	Yes
Freeze-thaw stability	Up to 10 cycles thawed at room temperature for both CT-P6 and US-licensed Herceptin	Yes
Frozen serum storage stability (days)	CT-P6: 35 days at -80 °C ± 10 °C and -25 °C ± 5 °C US-licensed Herceptin: 17 days at -80 °C ± 10 °C and -25 °C ± 5 °C	No**

*One additional QC each near LLOQ and ULOQ.

Long-term stability data has not been provided refer to **Table 6.

Performance of the validated ELISA method (ICD 546) during the sample analysis for study CT-P6 1.5 is summarized in **Table 6**.

Table 6. Summary of ELISA method performance in support of study CT-P6 1.5

^{(b) (4)} Project Code	RFHD2	Acceptability
Assay passing rate	• 87% of the assay met the method acceptance criteria.	Yes
Standard curve performance	• Cumulative bias range: -7.07 to 7.13% • Cumulative precision: ≤ 5%CV	Yes
QC performance	• Cumulative bias range: -0.7 to 8.6% • Cumulative precision: ≤ 13%CV • TE: ≤ 20%	Yes

Study sample analysis/ stability	Refer to Section 2.2 for the duration to establish serum storage stability.

Initially, serum trastuzumab concentrations from study CT-P6 1.5 were determined using a validated GyrosLab® -based immunoassay at (b) (4) (ICD 546). This method (M08.CT-P6 or Trastuzumab.huse.1) was developed at (b) (4) and transferred to (b) (4) where the validated range was further extended. Upon review of the submission, this method was found not suitable to accurately measure serum trastuzumab concentrations; see **Table 7** below for a summary of this method. In GyrosLab®-based immunoassay, biotinylated- antibody specific to the cytoplasmic domain of HER-2 were first allowed to pass through the microstructures containing streptavidin-coated beads in Bioaffy 1000 CDs, followed by full-length HER-2. Then, the serum samples containing trastuzumab were allowed to pass through these microstructures where trastuzumab then bound to HER-2. Immobilized trastuzumab was detected by Alexa Fluor® 647-conjugated purified mouse anti-human IgG1 antibody. Wash steps were performed between each reagent/ sample addition to remove any unbound material. The CD was read by a laser that excited the Alexa Fluor® 647, producing a fluorescent signal that was directly proportional to the concentration of trastuzumab. **Table 7** shows brief summary review explaining the deficiency of the method resulted in under-quantification of serum trastuzumab.

Table 7. Brief summary of GyrosLab® -based immunoassay method used to support study CT-P6 1.5

Bioanalytical method review summary	GyrosLab® -based immunoassay method had a deficiency since serum trastuzumab concentrations were not accurately measured within the validated calibration range due to following 2 main reasons; 1) the amount of capture reagents allowed to pass through the microstructures were limited compared to trastuzumab concentrations even after MRD, and 2) the presence of additional capture step (Capture 1 to capture 2) might require longer incubation time to reach an optimal equilibrium than GyrosLab® allowed.
(b) (4) Project code	RBNU2
method #	ICD 546
Material for calibration curve & concentration	CT-P6, Lot 13A3C005 at 21.0 mg/mL US-licensed Herceptin, Lot 554763, 21.0 mg/mL
Validated assay range	5.00 (LLOQ) – 750 (ULOQ) mcg/mL
Minimum required dilution (MRD)	1:1000 in sample diluent (filtered Superblock with 250 mcg/mL mouse IgG)
Source & lot of reagents	- Capture 1: Biotinylated- c-erbB-2/Her-2/neu Ab-17, clone e2-4001+3B5 (b) (4), Lot: NB#9145-67-01 - Capture 2: Recombinant Her-2 (b) (4), Lot: 111313 - Detection: Alexa Fluor® 647-conjugated purified mouse anti-human IgG (b) (4), Lot: NB#9145-84-01
Regression model & weighting	5-Parameter logistic, weighting of 1/Y ²

As a result of the inadequacies of the GyrosLab®-based immunoassay, 1) additional details regarding method validation are not included and 2) serum trastuzumab concentrations determined using a validated ELISA method described in **Table 5** were reviewed and used for the PK similarity assessment.

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/s/

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02/21/2018

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NAM ATIQR RAHMAN
02/21/2018
I concur with the recommendation.