This part outlines the scientific assessment and knowledge about this product at the time of prequalification. Updates to this information are included in parts 1 to 5 and 8 of this WHOPAR.

SCIENTIFIC DISCUSSION

Name of the Finished Pharmaceutical Product:	[BT-ON013 trade name][1]
Manufacturer of Prequalified Product:	Biocon Limited Special Economic Zone Plot Nos. 2, 3, 4 & 5 Phase-IV Bommasandra-Jigani Link Road Bommasandra Post Bengaluru – 560099, India
Drug Substance (DS):	Trastuzumab
Pharmaco-therapeutic group (ATC Code):	Antineoplastic agent, monoclonal antibody (L01XC03)
Therapeutic indication:	Treatment of early stage HER2 positive breast cancer or metastatic HER2 positive breast cancer

1. Introduction

[BT-ON013 trade name] contains trastuzumab, a humanized recombinant immunoglobulin G (IgG) monoclonal antibody, directed against the human epidermal growth factor receptor 2 (HER2). [BT-ON013 trade name] has been developed as a biosimilar for Herceptin (Roche).

[BT-ON013 trade name] has been prequalified for the treatment of metastatic breast cancer (MBC) and early breast cancer.

2. Assessment of quality

The assessment was done according to the "*WHO Pilot Procedure for Prequalification of Biotherapeutic Products: Rituximab and Trastuzumab*"[2], "*WHO Guidelines on evaluation of similar biotherapeutic products (SBPs)*"[3], "*WHO Questions and Answers: similar biotherapeutic products*"[4] "*WHO Guidelines on the quality, safety and efficacy of biotherapeutic protein products prepared by recombinant DNA technology*"[5] and other applicable guidelines.

[1] Trade names are not prequalified by WHO. This is the National Medicines Regulatory Authority's responsibility.
[2] https://www.who.int/medicines/regulation/biotherapeutic_products/en/
[3] Annex 2, Technical Report Series No. 977, 2009
[4] https://www.who.int/biologicals/expert_committee/QA_for_SBPs_ECBS_2018.pdf?ua=1
[5] Annex 4, Technical Report Series No. 987, 2014

Drug substance (DS)

General information

[BT-ON013] (trastuzumab) drug substance is a humanised monoclonal antibody of the IgG1 subclass. It is a glycoprotein with one N-linked glycosylation site on the Asn300. Trastuzumab selectively binds to the extracellular domain of HER2, thereby preventing HER2 signalling. The known mechanisms of action of trastuzumab are binding to HER2 leading to inhibition of cell proliferation, as well as target cell killing via antibody-dependent cell-mediated cytotoxicity (ADCC) activity.

Manufacturing process and process controls

The facility responsible for the manufacture and testing of the drug substance is Biocon Limited, Plot No. 2-5, Phase IV, Bommasandra-Jigani Link Road, Bommasandra post, Bengaluru, India. Trastuzumab DS is produced by recombinant DNA technology, being expressed in Chinese Hamster Ovary (CHO) cells. Following cell culture and harvest, the drug substance is purified from the harvest culture fluid through a series of filtration and chromatography steps. The process includes steps to inactivate/remove potential contaminating viruses. Excipients are added to generate the formulated drug substance.

The in-process controls (IPCs) performed at each stage during manufacture of the drug substance are acceptable. The definition of the critical process parameters (CPPs) and critical IPCs and justification for their limits are acceptable.

Control of raw materials

The raw materials are sufficiently described and controlled, with the stage of the manufacturing process where the material is used being provided. A two-tier cell banking system of Master Cell Bank (MCB) and Working Cell Bank (WCB) was established and qualified, in line with ICH Q5A and Q5D. Genetic stability up to and beyond the generation number needed for routine production was investigated.

Process validation

The process validation data demonstrate that the commercial process, when operated within the specified ranges, consistently produces drug substance that meets the predetermined specifications. Clearance of process-related impurities has been found to be consistent, with the impurity levels observed at the end of the manufacturing process sufficiently reduced to very low levels. Overall, the manufacturing process is considered appropriately validated. Chromatographic column resin reusability was evaluated.

Manufacturing process development

The manufacturing process for [BT-ON013 trade name] drug substance was scaled-up and revised over the course of development, and comparability studies between the scales demonstrated consistency of the DS.

Characterisation

A range of state-of-the-art orthogonal methodologies was used in order to elucidate the primary and higher order structures, as well as the purity/impurity, charged variants and glycan structures. Multiple biological assays were employed addressing multiple putative mechanisms of action of trastuzumab, in order to assess the potency and binding affinity of [BT-ON013 trade name] DS. The results demonstrate that [BT-ON013 trade name] DS has the expected structure and functional properties. (See Table 1, under "Assessment of Analytical Comparability" for list of analytical procedures used in characterization of Biocon's trastuzumab).

Specifications

Specifications are set in accordance with ICH Q6B and include tests for appearance, identity, purity and impurities, content, potency, microbiological safety and general tests. The specifications are considered sufficient to control the quality of the drug substance.

The analytical methods used for active substance testing and their validation have been described in detail. The methods are appropriately validated.

Reference materials

The information provided on present and past reference standards is considered sufficient.

Container closure system

[BT-ON013 trade name] formulated DS is stored in suitable packaging that is compliant with relevant compendial monographs and other accepted quality standards.

Stability

The stability data support the proposed shelf-life of 24 months, when the active substance is stored under the recommended long-term storage conditions.

Drug Product (DP)

Product description

[BT-ON013 trade name] is a sterile, off-white to pale yellow lyophilized powder, which upon reconstitution with sterile water for injection (WFI), yields a single-dose, colorless to pale yellow transparent solution, free of visible particles.

The excipients of [BT-ON013 trade name] are L-histidine hydrochloride, L-histidine, sorbitol, macrogol 3350, hydrochloric acid (for pH adjustment) and sodium hydroxide (for pH adjustment).

[BT-ON013 trade name] is supplied as a single-use vial containing 150 mg of trastuzumab as drug substance. The DP packaging consists of a clear, colourless Type I glass vial, closed with a 20 mm chlorobutyl rubber stopper with fluoro-polymer laminate on the product contact side. The rubber stopper is sealed with an aluminium seal that has a plastic flip-off cap.

Pharmaceutical development

Biocon focused on developing a trastuzumab DP formulation that was highly similar to the reference product, Herceptin® (manufactured by Roche), from a quality and stability perspective.

The changes made to the manufacturing process during the development have been made to improve control of the process, with no impact to the quality of the final drug product. The comparative analytical data of the finished product used in the manufacture of Phase I, Phase III and process validation batches were provided.

The container closure system was described and adequately qualified; leachables and extractables studies were performed.

Manufacture of the product and process control

The Biocon's trastuzumab 150 mg DP manufacturing process involves thawing of formulated DS, pooling of individual DS bags followed by mixing, pre-filtration, sterile filtration, aseptic filling of the formulated DS, lyophilization, and sealing of vials containing the lyophilized product. No additional excipients are added during the preparation of the finished medicinal product, as excipients are added during the active substance manufacturing process. Critical process parameters have been identified and are acceptable. In-process controls are appropriately justified and are performed at each stage during the manufacture of the finished medicinal product.

The manufacturing process of Biocon's trastuzumab 150 mg DP was validated using three consecutive full-scale production batches. The process has been adequately validated and the data provided indicate that the process is under control and yields a product meeting the pre-defined quality characteristics.

The aseptic process used for the sterilization has been validated through media fills.

Shipping

Procedures are in place in order to ensure that the containers are maintained at the required temperature throughout the shipment duration, from origin to destination. The containers are transported from the origin in a temperature-controlled truck, shipped through an airline and stored in the airline's temperature-controlled facility upon arrival and finally shipped to destination using temperature-controlled trucks.

The shipping validation for [BT-ON013 trade name] was evaluated, by simulation of the mode of transportation and conditions adopted for the transportation of the drug product during transit using the worst route with halt. The studies presented are sufficient to ensure that the temperature control system can maintain the required temperature from origin to destination. Acceptability of short-term excursions outside the labelled storage conditions are supported by temperature excursion/thermal cycling studies conducted for Biocon's trastuzumab DP. Any temperature excursions received from customers will be investigated and assessed for the impact on product quality according to relevant standard procedures. Investigation details and assessment outcome are further notified to the customer.

Every shipment should include a temperature-monitoring device meeting the minimum specifications outlined in the WHO-applicable guidelines.

Product specifications

The release specification for the finished product includes tests for appearance, identity, purity and impurities, content, excipient, potency, pH, general pharmacopoeial tests and safety testing. The drug product specifications are considered adequate and in accordance with ICH Q6B.

The analytical methods used for routine testing of the finished product have been appropriately described and non-compendial methods have been validated in accordance with ICH Q2 (R1).

Stability of the finished product

Stability studies under long-term, accelerated and stress storage conditions, have been performed for [BT-ON013 trade name] batches according to ICH Q5C guideline. Stability studies were carried out on six commercial scale batches of Biocon's trastuzumab DP. The quality attributes selected for the stability studies are considered adequate to evaluate the stability of trastuzumab DP. All the results were within the predefined specifications.

An in-use stability study was performed and supports the stability of the product reconstituted as indicated in the SmPC.

An infusion study showed compatibility with infusion bags/systems of polyvinyl chloride (PVC), polypropylene (PP) or polyethylene (PE) materials at 30°C for a period of 24 hours.

A temperature excursion study showed that the product was stable after being exposed to a temperature excursion (25°C) of 48 hours

The proposed shelf life of 48 months for [BT-ON013 trade name] when stored at 5°C±3°C in the container closure system intended for commercial use is supported.

3. Assessment of Analytical Comparability

A comprehensive exercise has been conducted to demonstrate similarity of Biocon's trastuzumab DP to the reference products (US-approved and EU-approved Herceptin, by Roche). A step-wise approach using extensive, state-of-the-art analytical techniques to characterize the physicochemical and biological properties of Biocon's trastuzumab and the reference products has been used.

The analytical similarity of Biocon's trastuzumab DP, and the reference product was evaluated using sensitive techniques to examine primary, secondary and tertiary structure, content, impurities, charge variants, glycosylated variants and other post-translational modifications. In addition, extensive evaluation of HER-2 binding, Fc binding and resulting biological effects such as ADCC have been compared. Analytical similarity of Biocon's trastuzumab, and the reference product was assessed using multiple methods, many of which are orthogonal. The methods were validated or qualified at the time of testing and established as suitable for intended use.

Table 1: Quality & Biological Attributes and Methods Used to Evaluate the Analytical Similarity of Biocon's Trastuzumab and Herceptin

Quality/Biological Attribute	**Methods**
Primary structure	Peptide mass fingerprinting by liquid chromatography (LC) with electrospray (ESI) mass spectrometry (MS) detection
	Intact molecular mass (LC-ESI-MS)
	Reduced molecular mass (LC-ESI-MS)
Protein content	Concentration [ultraviolet (UV) spectroscopy at 280 nm]
Higher order structure	LC-ESI-MS (disulfide bond characterization)
	Fourier transform infrared spectroscopy (FTIR)
	Ellman's reagent (free cysteines)
	Near and far UV circular dichroism (CD)
	Differential scanning calorimetry (DSC)
	Intrinsic fluorescence (IF)
Size Variants (including aggregates)	Size exclusion high performance liquid chromatography (SEC-HPLC) with UV detection
	SEC with multi-angle light scattering (MALS)
	Capillary Electrophoresis-Sodium Dodecyl Sulfate (CE-SDS, Non-Reduced)
	Analytical ultracentrifugation sedimentation velocity
Charge and hydrophobic variants	LC-ESI-MS (oxidation)
	Capillary isoelectric focusing (cIEF)
	Cation exchange HPLC (CEX-HPLC)
	Hydrophobic interaction chromatography (HIC-HPLC)
Glycosylation	Boronate affinity chromatography (glycation)
	Reverse phase -HPLC (afucosylation and sialic acid)
	CE-SDS reduced (Non-glycosylated heavy chain)
	Normal phase (NP)-HPLC (glycan mapping)
Oxidation at Met255	Reverse phase –HPLC with ESI-MS
Sub-visible particles	Micro-flow imaging (MFI)
Potency	Antibody-dependent cellular cytotoxicity (ADCC) bioassay using peripheral blood mononuclear cells **(**PBMC)
	HER2 binding assay by flow cytometry
	Inhibition of proliferation bioassay
Fc-receptor binding and function	Cellular dependent cytotoxicity (CDC) bioassay
	C1q binding assay (ELISA)
	FcRn binding affinity (Biacore)
	FcγRIa binding affinity (Biacore)
	FcγRIIa binding affinity (Biacore)
	FcγRIIb binding affinity (Biacore)

	FcγRIIIa V type binding affinity (Biacore)
	FcγRIIIb binding affinity (Biacore)

All the analyzed lots of the reference product were within their respective shelf life. Multiple lots of the reference product with a remaining shelf life between 5 and 48 months have been analyzed using various analytical methods. Biocon's trastuzumab batches used in the analytical similarity assessment span an age of 1 month to 43 months. Biocon's trastuzumab batches are derived from different drug substance batches in order to account for variability in the manufacturing process of the drug substance.

Most of the quality attributes proved to be highly similar between Biocon's trastuzumab and the reference product. Nevertheless, for some structural parameters, which might impact clinical performance, differences were observed and are discussed below.

Levels of non-glycosylated heavy chain have been found to be lower in Biocon's trastuzumab compared to the reference product, but the difference was found to be very small and it is not expected to have any impact on the product's biological activity.

The total high mannose content for Biocon's trastuzumab was found to be higher but within the range of the reference product. The *in vitro* bioactivity assay data dependent on Fc function (ADCC and FcγRIIIa) indicate similarity with the batches of the reference product.

Afucosylated content for Biocon's trastuzumab is slightly higher compared to the reference product. However, the *in vitro* bioactivity assay data dependent on Fc function (ADCC and FcγRIIIa) indicate similarity of the two products.

The slight differences in sialic acid content are not considered to have an impact on pharmacokinetics and biological activity due to the low overall content.

Taken together, the small differences observed between test and reference products have been appropriately justified and the data provided indicate that [BT-ON013 trade name] can be considered as biosimilar to the reference product with respect to quality.

Arrangement for handling complaints and recalls

The Applicant provides detailed descriptions of the arrangements for handling complaints and recalls, including manufacturer-initiated recalls, and of the distribution process. The procedures have been found to be sufficient.

Conclusions

The quality part of the dossier is accepted.

4. Assessment of non-clinical data

Overview

The nonclinical program conducted by Biocon included a comparative single-dose PK study in female cynomolgus monkeys and a comparative repeat-dose toxicity study with 5 weekly

administrations of test articles in male and female cynomolgus monkeys. The reference product Herceptin was used in both studies. The repeat-dose study was performed under GLP norms as described by the Organisation for Economic Co-operation and Development, and based on ICH S6 (R1). Additionally, *in vitro* studies were performed in adult human and neonatal rat cardiomyocytes to examine the relative cardiotoxicity of Herceptin and Biocon's trastuzumab.

The initial formulation was developed to have the same composition as that of intravenous Herceptin. This formulation was referred to as the Bmab 200-reference product formulation (Bmab 200-RPF), however, macrogol 3350/PEG 3350 was then selected as an alternative cryoprotectant and D-sorbitol was selected as a lyoprotectant and bulking agent. The resulting formulation is proposed for prequalification.

Pharmacology

Primary Pharmacodynamics

The comparative analysis of Biocon's trastuzumab and the reference product showed that ADCC activity and FcγRIIIa binding kinetics (K_a, K_d and K_D) of the two products were similar. Furthermore, the study results showed no significant difference in the K_a, K_d and K_D between Biocon's trastuzumab and the reference product with regards to: Fc-receptor binding (FcRn, FcγRIa [CD64], FcγRIIa [CD32A], cRIIb [CD32B], FcγRIIIb); affinity to complement (C1q); complement-dependent cytotoxicity (CDC) activity; inhibition of the proliferation of the SK-BR-3 cells; and HER2 receptor binding. (SKBR3 cells are human breast cancer cells that overexpress HER2).

Secondary pharmacodynamics

The applicant did not present any secondary pharmacodynamic data on the basis that it is not required for the prequalification of a biosimilar medicinal product. This was acceptable.

Safety pharmacology

The studies did not identify relevant cardiotoxicity in the repeated dose toxicology study.

The comparison in cardiotoxicity performed *in vitro* in rat and human cardiomyocytes revealed no differences between the reference product and Biocon's trastuzumab.

Pharmacodynamic drug interactions

Biocon Limited did not perform any studies to examine pharmacodynamic drug interactions as Biocon's trastuzumab was claimed to be a biosimilar product. Several pharmacodynamic drug interactions studies present in the public domain were described by the applicant in the appropriate sections of the dossier.

Overall conclusions on pharmacology

The reported studies did not show significant differences between the reference product and Biocon's trastuzumab.

Pharmacokinetics

The pharmacokinetics (PK) of Biocon's trastuzumab and the reference product were determined in cynomolgus monkeys after a single 30-minute intravenous (IV) dose.

Information on distribution, metabolism, excretion and pharmacokinetic interactions were not provided but those studies are not required for a biosimilar medicinal product.

Toxicology

The toxicology program for Biocon's trastuzumab consisted of one pivotal, GLP-compliant, two-way comparative repeat-dose toxicity study, performed in cynomolgus monkeys, administered weekly 25 mg/kg or 50 mg/kg IV on 5 occasions for 4 weeks. The toxicokinetic results indicated there were no notable differences in Biocon's trastuzumab's and the reference product's exposure or bioavailability to monkeys.

Single dose toxicity study, reproductive and developmental, carcinogenicity, genotoxicity studies were not performed. This is considered acceptable for an application for a biosimilar product.

No specific local tolerance studies were conducted, but tolerance was evaluated in the repeat-dose toxicity study. In the single-dose study, erythema and desquamation were reported at the injection sites, with similar frequency and severity for Biocon's trastuzumab and reference product. In the repeat-dose study, no signs of erythema, oedema, atonia, desquamation, or fissuring were evident in any animal.

Conclusion on the non-clinical aspects

Overall, the nonclinical data indicate that Biocon's trastuzumab has an activity similar to that of the reference product with an acceptable safety profile. The non-clinical part is accepted.

5. Assessment of Clinical Data

The Clinical trials were performed in accordance with GCP as claimed by the applicant.

Summary of clinical studies

Type of Study	Study Number	Study Objective(s)	Study Design	Test Product(s), Dosage, Regimen, Route of Administration	Number of Subjects/ Diagnosis	Duration of Treatment
Pivotal Studies						
PK bioequiv-alence, PD, safety, immuno-genicity	MYL-Her-1001	• To confirm PK bioequivalence between MYL-1401O and EU-Herceptin® • To assess comparative safety and tolerability • To investigate PD parameters	Single-center, single-dose, 2-period, double-blind, crossover study	MYL-1401O, EU-Herceptin 8 mg/kg single dose IV	22 randomized, 19 completed/ Healthy male subjects	Single IV dose administered over 90 min
PK, safety, immuno-genicity	MYL-Her-1002	• To demonstrate PK similarity of MYL-1401O vs EU-Herceptin and US-Herceptin along with EU-Herceptin vs US-Herceptin • To further assess	Single-center, single-dose, randomized, double-blind, 3-arm, parallel-group study	MYL-1401O, EU-Herceptin, US-Herceptin 8 mg/kg single dose IV	132 randomized, 121 completed/ Healthy male subjects	Single IV dose administered over 90 min

Type of Study	Study Number	Study Objective(s)	Study Design	Test Product(s), Dosage, Regimen, Route of Administration	Number of Subjects/ Diagnosis	Duration of Treatment
		similarity of PK among MYL-1401O, EU-Herceptin, and US-Herceptin • To assess comparative safety				
Confirmatory efficacy and safety, immuno-genicity	MYL-Her-3001	• To compare the independently assessed best ORR at Week 24 • To compare independently assessed clinical activity at Week 24 (TTP, PFS, OS) • To descriptively compare safety, tolerability, and immunogenicity • To compare population PK • To assess impact of shed ECD fragments on HER2 receptor on PK and efficacy parameters	Multicenter, double-blind, randomized, parallel-group study	MYL-1401O, EU-Herceptin 8 mg/kg loading dose followed by 6 mg/kg maintenance, every 3 weeks for 8 cycles IV	500 randomized, 356 completed Part 1/ Patients with HER2+ MBC	48 weeks
Supportive Study						
PK, comparative efficacy and safety, immuno-genicity	BM200-CT3-001-11	• To evaluate and compare the single-dose PK parameters of Bmab-200 and EU-Herceptin • To evaluate and compare ORR • To evaluate and compare the multi-dose PK • To assess comparative safety and immunogenicity • To correlate secondary efficacy parameters with shed HER2 ECD	Multicenter, double-blind, randomized, parallel-group study	Bmab-200, EU-Herceptin 8 mg/kg loading dose followed by 6 mg/kg maintenance, every 3 weeks for 8 cycles IV	135 randomized, 103 completed /Patients with HER2+ MBC	24 weeks

Pharmacokinetics (PK)

Pivotal pharmacokinetic data for Biocon's trastuzumab were available from 2 studies in healthy volunteers: MYL-Her-1001 and MYL-Her-1002.

A population PK analysis was also conducted in patients with HER-2+ metastatic breast cancer in study MYL-Her-3001. Supportive PK data are also provided from study BM200-CT3-001-11.

Pivotal PK studies

Study MYL-Her-1001 was a single-center, single-dose, two-period, randomized, double-blind, crossover study in healthy male volunteers.

The study duration was up to 39 weeks, which included a screening period of 3 weeks, an initial randomization and treatment Period 1 of 14 weeks, an interim (washout) period of up to 8 weeks (mean 5 weeks), and a crossover treatment Period 2 of 14 weeks. During each study period, 18 blood samples for PK testing were collected from each subject at 0, 45, and 90 minutes (just prior to the end of infusion); at 3, 6, 9, 24, 48, and 96 hours post-dose (relative to the start of infusion); and at 8, 11, 15, 22, 29, 43, 57, 71, and 99 days post-dose. The PK parameters were determined using non-compartmental analyses, with C_{max} and $AUC_{0-\infty}$ as the co-primary endpoints.

Nineteen subjects were enrolled in this study. The subjects either received Biocon's Trastuzumab (MYL-1401O) or EU-approved Herceptin in Period I and an alternative treatment in Period II.

Primary objective: to ascertain pharmacokinetic (PK) bioequivalence of Biocon's Trastuzumab and Herceptin® after a single dose of 8 mg/kg was infused intravenously over 90 minutes in healthy male subjects. All primary PK analyses were based on the per protocol (PP) set. Bioequivalence was established if the 90% confidence interval of the mean ratio of Biocon's Trastuzumab to Herceptin met the standard bioequivalence criteria of 80-125% for the area under the serum concentration curve from 0 to infinity ($AUC_{0-\infty}$) and maximum observed serum concentration (C_{max}).

Secondary objectives: to assess comparative systemic safety and tolerability, including local tolerance, and to evaluate immunogenicity with anti-drug antibody (ADA) formation.

• *Primary PK endpoints:*

Normalized $AUC_{0-\infty}$ and C_{max} (normalized to 8.0 mg/kg dose), relative bioequivalence bioavailability of Bicon's trastuzumab to Herceptin®

• *Secondary PK endpoints:*

Native (not normalized) $AUC_{0-\infty}$, native C_{max}, native AUC from zero to time of last quantifiable concentration (AUC_{0-last}), percentage of the $AUC_{0-\infty}$ extrapolated to infinity (AUC%extrap), time to reach maximum serum drug concentration (t_{max}), terminal elimination half-life ($t_{1/2}$), volume of distribution (V_z), volume of distribution when extrapolated to steady state (V_{SS}), total serum clearance (CL).

Pharmacokinetic results

In total, 22 subjects were randomized, with 22 receiving Herceptin and 19 receiving Biocon's Trastuzumab (MYL-1401O). The per protocol (PP) population for bioequivalence consisted of 19 subjects exposed to both drugs. Three subjects dropped out of the study after Period I (all after receiving Herceptin): 2 due to personal reasons (1 after 14 weeks and 1 after 8 weeks) and 1 due to transaminase elevation.

Table: Primary and secondary PK parameters as geometric means (GeoCV%) (PP population)

Parameter	Units	Hercules N=19	Herceptin® N=19	PE (90% CI)
Primary parameters				
C_{max} normalized	µg/mL	165 (15.7)	178 (15.6)	0.9218 (0.8760 ; 0.9699)
$AUC_{0-\infty}$ normalized	µg.h/mL	45486 (22.7)	48350 (28.5)	0.9368 (0.8874 ; 0.9889)
Secondary parameters				
C_{max} native	µg/mL	167 (14.7)	175 (15.8)	0.9417 (0.8997 ; 0.9858)
$AUC_{0-\infty}$ native	µg.h/mL	45802 (23.0)	47547 (28.6)	0.9571 (0.9048 ; 1.0123)
T_{max} [a]	h	1.5 (1.4-9.0)	1.5 (1.3-9.0)	
$T_{1/2}$ (day)	Day	6.94 (22.6)	7.02 (26.3)	0.9880 (0.9428 ; 1.0353)
V_z [b]	L	2.96 (18.0)	2.81 (18.0)	1.0547 (1.0126 ; 1.0985)
V_{ss} [b]	L	4.38 (17.6)	4.30 (15.1)	1.0190 (0.9681 ; 1.0726)
CL [b]	L/day	0.296 (22.7)	0.278 (28.5)	1.0675 (1.0112 ; 1.1269)

Normalized $AUC_{0-\infty}$ = Area under the serum concentration-time curve from time zero to infinity (normalized to a dose of 8.0 mg/kg); Normalized C_{max} = Maximum observed serum concentration (normalized to a dose of 8.0 mg/kg); CI = Confidence interval; CL = Total serum clearance; CV = Coefficient of variation; N = Number of subjects; PE = Point estimate; PK = Pharmacokinetic; PP = Per-protocol.

[a] T_{max} reported as median and range (no formal statistical analysis)

[b] Parameters adapted to a 70 kg body weight

Point estimate as ratio of geometric means Hercules vs. Herceptin® (difference of adjusted means after back transformation)

Study MYL-Her-1002 was a single-center, single-dose, randomized, double-blind, 3-arm, parallel-group study investigating the bioequivalence of Biocon's Trastuzumab (MYL-1401O) versus EU-approved Herceptin and US-licensed Herceptin as well as EU-approved Herceptin versus US-licensed Herceptin after a single dose of 8 mg/kg was administered as IV infusion over 90 minutes in healthy male subjects, under fasting conditions. One hundred thirty-two (132) volunteers were enrolled in the study.

Primary objective: to demonstrate pharmacokinetic similarity of Biocon's trastuzumab (Hercules) versus EU-approved Herceptin® and US-licensed Herceptin®, and pharmacokinetic similarity of EU-approved Herceptin® versus US-licensed Herceptin®.

Endpoint: The 90% confidence interval for the LSMeans ratio of C_{max}, AUC_{0-last}, and $AUC_{0-\infty}$ for the test and reference products was set between 80% and 125% for the natural log-transformed data.

The design of the pivotal PK studies was found to be acceptable and in line with relevant guidelines including the WHO '*Guidelines on evaluation of monoclonal antibodies as similar biotherapeutic products (SBPs)*'[3].

The analytical methods used were adequately validated and are acceptable.

Pharmacokinetic results

In total, 121 subjects completed the clinical portion of the study, but only 120 subjects were included in the PK analysis due to 1 subject receiving an incorrect dose of study medication resulting from a dose preparation error.

The results demonstrated that Biocon's Trastuzumab is bioequivalent to EU-Herceptin and US-Herceptin, and that EU-Herceptin is bioequivalent to US-Herceptin, as the 90% CIs for the natural log-transformed parameters, $LNAUC_{0\text{-last}}$, $LNAUC_{0\text{-}\infty}$, and LNC_{max} for trastuzumab were within 80% to 125% for the test to reference ratio.

Table: Mean (%CV) dose-normalised PK parameters – Biocon's trastuzumab vs EU Herceptin

Hercules vs. Herceptin EU Trastuzumab				
Parameter	**Arithmetic Mean (%CV) A = Hercules N=42**	**Arithmetic Mean (%CV) B=Herceptin-EU N=41**	**LSMEANS Ratio (A/B)***	**90% Confidence Interval****
$AUC_{0\text{-last}}$ (mcg•hr/mL)	48055 (15.92)	49823 (19.61)	0.97	91.31% – 103.05%
$AUC_{0\text{-}\infty}$ (mcg•hr/mL)	48241 (16.19)	50075 (19.81)	0.97	91.17% – 102.97%
C_{max} (mcg/mL)	200.4 (12.34)	192.6 (14.13)	1.04	99.00% – 109.82%
λ_z (hr^{-1})	0.0046 (22.80)	0.0044 (27.14)		
$t_{½}$ (hr)	160.0 (28.39)	173.8 (32.92)		
t_{max} (hr)	2.880 (54.83)	3.028 (118.2)		

*Ratio (A/B) = $e^{[LSMEANS\ of\ (LNA-LNB)]}$
**Used Natural Log Transformed Parameter

Table: Mean (%CV) dose-normalised PK parameters – Biocon's trastuzumab vs US Herceptin

Trastuzumab Hercules vs. Herceptin US Trastuzumab				
Parameter	**Arithmetic Mean (%CV) A = Hercules N=42**	**Arithmetic Mean (%CV) C=Herceptin-US N=37**	**LSMEANS Ratio (A/C)***	**90% Confidence Interval****
$AUC_{0\text{-}last}$ (mcg•hr/mL)	48055 (15.92)	49826 (13.98)	0.96	90.34% – 102.29%
$AUC_{0\text{-}\infty}$ (mcg•hr/mL)	48241 (16.19)	50181 (13.86)	0.96	89.96% – 101.94%
C_{max} (mcg/mL)	200.4 (12.34)	197.9 (16.25)	1.02	96.42% – 107.26%
λ_z (hr^{-1})	0.0046 (22.80)	0.0042 (23.45)		
$t_{½}$ (hr)	160.0 (28.39)	176.4 (29.85)		
t_{max} (hr)	2.880 (54.83)	2.625 (53.37)		

*Ratio (A/C) = $e^{[LSMEANS\ of\ (LNA-LNC)]}$
**Used Natural Log Transformed Parameter

The primary PK analysis showed PK comparability of the test and reference product at the dose of 8mg/kg body weight. The 90% confidence intervals were well contained within the pre-specified standard bioequivalence margins of 80-125%. The secondary endpoints were also similar across the treatment groups.

Supportive PK study

Study BM200-CT3-001-11

This was a double-blind, randomized, active-controlled, parallel-group, comparative study in patients with HER2-positive metastatic breast cancer to evaluate the comparative PK, efficacy, safety, and immunogenicity of Biocon's trastuzumab with EU-approved Herceptin. This study was conducted to meet the requirements for marketing authorization in the country of origin (India) and the formulation used (Bmab-200) was slightly different from that used in the pivotal studies.

The 90% confidence intervals for the ratios of both primary endpoints C_{max} and $AUC_{0\text{-}t}$ were well contained within the conventional 80%-125% interval, supporting biosimilarity.

Pharmacokinetics in the target population

In addition to the pivotal PK data collected in healthy volunteers, supportive PK data were also provided in the target population from a population PK analysis undertaken in study **MYL-Her-3001**.

Part 1 of MYL-Her-3001 study was the main part for demonstration of efficacy as well as PK assessment, including 8 cycles (24 weeks) of combination therapy. The PK population included all randomly assigned patients who received at least 1 complete dose of Biocon's Trastuzumab or Herceptin, and who provided at least 1 post-dose sample for PK analysis.

Model development included assessment of covariate effects on the inter-individual variability in PK parameters. A bootstrap analysis and goodness-of-fit plots, including visual predictive checks, were presented to evaluate the robustness of the final model. Observed C_{min} values at the end of Cycle 1 and Cycle 6 were used to assess the similarity of Biocon's Trastuzumab versus Herceptin using the two 1-sided t-tests statistical approach for bioequivalence. Individual patient empiric Bayesian parameter estimates were used to estimate PK measures reflecting exposure to drug and were compared qualitatively between treatments.

The results showed that the population PK profiles of Biocon's trastuzumab versus EU-approved Herceptin were not different in patients with HER2-positive MBC. Treatment was not a significant covariate of clearance (p=0.176) or volume of the central compartment (p=0.567) using the likelihood ratio Chi-square test. The model showed that observed trough concentrations were not different between treatments at the end of the first dosing interval or at Cycle 6 supporting biosimilarity.

Special populations: analyses in special populations were not submitted but are not relevant in the Biocon's trastuzumab as the biosimilar product relies on the information already known of the reference product. No formal drug-drug interaction studies are needed.

Overall conclusions on pharmacokinetics

The data provided is adequate and supports biosimilarity of Biocon's trastuzumab and the reference product.

Pharmacodynamics and Pharmacokinetics-Pharmacodynamics (PK/PD)

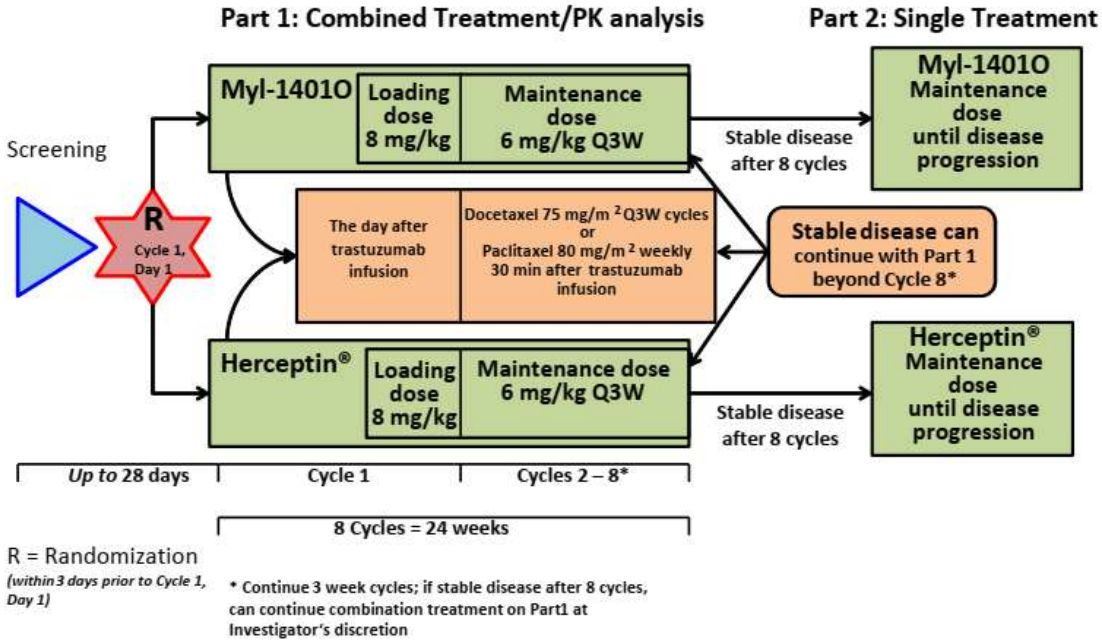
Currently no PD markers have been validated for prediction of efficacy of trastuzumab. However, PD was evaluated in Study MYL-Her-1001 as an exploratory objective to support the biosimilarity assessment of Biocon's Trastuzumab and Herceptin. Immunomodulation was assessed using cytokine production in serum, mononuclear cell subset immunomodulation, and stimulation of peripheral blood mononuclear cells to measure cytokine production. Cytotoxicity was assessed using markers of apoptosis in PBMCs (caspase-3 activation, DNA fragmentation) and Akt phosphorylation, and anti-proliferative activity (proliferation inhibition) of ex vivo serum samples on a cell line overexpressing HER2 receptors. There were no significant differences seen between Biocon's trastuzumab and the reference product for any of the PD parameters analysed, although many showed marked changes over time for both groups. This data was exploratory and the pivotal data to support biosimilarity for clinical pharmacology was derived from the PK data from clinical studies MYL-Her-1001 and 1002. However, these PD results, together with the PK data, supported biosimilarity between Biocon's trastuzumab and the reference product.

Clinical efficacy

The pivotal confirmatory efficacy and safety study (**MYL-Her-3001**) aimed to evaluate biosimilarity between Biocon's trastuzumab (Myl-1401O) and the reference product. The study consists of two parts:

- Part 1: the main part for demonstration of efficacy as well as PK assessment including 8 cycles (24 weeks) of combination therapy
- Part 2: evaluating safety and immunogenicity of Biocon's trastuzumab (Myl-1401O) compared with the reference product.

Figure 1 Study Design



PK: pharmacokinetic; Q3W: every 3 weeks; R: randomization.
Source: Study Protocol (Appendix 16.1.1).

Both drugs were administered as a single agent until disease progression. It should be highlighted that patients were allowed to continue concomitant taxane treatment if it was the investigator's opinion that they would benefit from it.

Duration of Part 1 and Part 2 of the study were selected to approximately correspond to the median progression free survival (PFS) for first-line treated MBC patients. The overall duration of the study was selected to correspond with the median overall survival (OS) for first-line treated MBC, which is considered acceptable. The primary endpoint was best overall response rate (ORR) at week 24, and was considered objective and sensitive for detecting differences between the test product, Biocon's trastuzumab and the reference product. At 24 weeks, this correlates with important clinical outcomes (PFS or time to progression [TTP]), as seen in the pivotal trial of the reference product.

Inclusion and exclusion criteria were adequate, reflecting a patient population sufficiently sensitive for showing biosimilarity between Biocon's trastuzumab and the reference product.

The main inclusion criteria were:

- Adult subjects with histologically confirmed diagnosis of breast cancer, locally recurrent or MBC that was not amenable to curative surgery and/or radiation.
- Documentation of HER2 gene amplification by fluorescent in situ hybridization (FISH) (as defined by a ratio > 2.0) or documentation of HER2-overexpression by immunohistochemistry (IHC) (defined as IHC3+, or IHC2+ with FISH
- Documentation of ER/PgR status (positive or negative) based on either a local or central laboratory report must have been available before randomization.
- Pathologically confirmed breast cancer with at least 1 measurable metastatic target lesion (based on RECIST 1.1 criteria). Bone, central nervous system (CNS), and skin lesions, as well as lesions that were irradiated, biopsied, or had any form of local intervention or surgical manipulation, were only to be assessed as non-target lesions.
- Patients previously treated with trastuzumab or lapatinib in the adjuvant setting were allowed if metastatic disease was diagnosed at least 1 year after the last dose
- Prior treatment with hormonal agents or bisphosphonates/denosumab was allowed.
- ECOG PS of 0 to 2.

Main exclusion criteria:

- Prior systemic therapy in the metastatic disease setting. This included: chemotherapy, signal transduction inhibitors (e.g., lapatinib), HER2 targeted therapy (e.g., trastuzumab), or other investigational anticancer therapy.
- Prior treatment with neoadjuvant or adjuvant anthracyclines with a cumulative dose of doxorubicin of > 400 mg/m^2 or epirubicin of > 800 mg/m^2.

It should be highlighted that inclusion of patients receiving trastuzumab as both first- and second-line treatment for MBC was initially allowed. Forty-two patients were randomized. Following a protocol amendment in view of including a more homogeneous population and to better reflect clinical practice, inclusion was limited only to patients receiving trastuzumab as first-line treatment for MBC.

Analysis sets

The ITT Population #1 (ITT1) consisted of all patients who were randomized into the study under Protocol Amendment 2 (Version 5.0; 11 Oct 2013) and beyond. Patients in ITT1 were categorized to the treatment as randomized. The primary efficacy analysis was conducted in ITT1.

The ITT Population #2 (ITT2) consisted of all randomized patients (i.e., included patients enrolled under Protocol Amendment 1 [Version 2.0; 02 Jul 2012]). Protocol Amendment 1, which was the first version of the protocol under which patients were enrolled, allowed randomization of patients who would receive second-line treatment for MBC (42 patients were randomized under Protocol Amendment 1). Patients randomized under Protocol Amendment 1 were included in the ITT2 but excluded from the ITT1 population The ITT2 population consisted of all randomized patients.

PP population: The PP population was defined at the end of Part 1 and was a subset of patients in the ITT1 who met the following criteria:
-Received the treatment to which they were randomized,
-The absence of any major protocol violations in Part 1 which precluded the evaluation of the patient including, for example, the lack of measurability of the lesions; the absence of violation of entry criteria which completely precluded the assessment of efficacy and safety. The details of all major protocol deviations that removed patients from the PP population were described in the BDRM Highlights, (see

details in the conduct of the study paragraph)
-Had at least 1 post-baseline tumor assessment if a progression disease; and at least 2 if CR, PR, or SD,
-Had received at least 2 complete cycles of treatment; however, if a progression, death, or discontinuation occurred before the end of the first 2 cycles, the patient was retained in the PP population.

826 patients were screened, 326 were screening failures (most common reason being lack of HER2+ confirmation). 500 patients were randomized (**458 according to amendment 2 is the ITT1 population**); considering the ITT1 230 were assigned to the MYL-1401O arm, 173 completed Part 1 of the study, while 228 randomized to the Herceptin2 arm, 159 completing part 1. 55 and 65 patients respectively in the MYL-1401O arm and Herceptin arm discontinued phase 1 of the study, the main reason being disease progression which occurred in a slightly higher percentage of subjects treated with Herceptin (17.9% versus 22.8%).

Considering the safety population: 342 participate in part 2 (179 MYL-1401O and 163 Herceptin patients), of these subjects a small group 15 and 17 in the MYL-1401O and Herceptin arm respectively, continued taxane. Similar number of patients (63 and 65 of MYL-1401O and Herceptin arm, respectively) discontinued part 2 of the study, disease progression occurred in a very similar percentage of subjects i.e. 31.3% and 31.9% in MYL-1401O and Herceptin arm, respectively.The treatment combination trastuzumab (Biocon's trastuzumab/Herceptin) plus a taxane (docetaxel or paclitaxel) is considered in line with clinical practice in settings where pertuzumab is not available. Pertuzumab is a HER2/neu receptor antagonist indicated in combination with trastuzumab and docetaxel for treatment of patients with HER2-positive MBC who have not received prior anti-HER2 therapy or chemotherapy for metastatic disease.

Dosing of trastuzumab corresponded to that approved in the EU SmPC of Herceptin for MBC i.e. 8 mg/kg loading dose on Cycle 1, Day 1, by continuous intravenous infusion over 90 minutes (min), and the maintenance dose was 6 mg/kg trastuzumab by continuous intravenous infusion over 30 minutes (±10 min) every 3 weeks. The selected dose regimen for docetaxel, a dose of 75 mg/m^2 of Body Surface Area (BSA) administered intravenously over 1 hour (±10 minutes) every 3 weeks throughout the study was acceptable. A dose range of 30 to 100 mg/m^2 has been used in trials as well as in practice. The 75mg/m^2 dose is however, the most widely used as higher doses are associated with greater toxicity.
The selected dose regimen for paclitaxel (weekly schedule of 80 mg/m^2) was acceptable and based on results from clinical trials showing better efficacy of the weekly schedule versus every 3 weeks.

*Primary Endpoint***:** best ORR at week 24.
Objective response was defined as a complete response (CR) or partial response (PR) according to RECIST 1.1 criteria, based on central tumor evaluation (taking as reference for PD the smallest measurements recorded since the treatment started).

Best ORR was considered a more sensitive endpoint than PFS, hence ORR was considered an appropriate primary endpoint. The assessment at 24 weeks was chosen based on results from Cleopatra study showing at this time point a significant difference in ORR between Herceptin + docetaxel and docetaxel alone and is considered appropriate.
The equivalence analysis was based on the ratio of the ORRs as (primary analysis) and on the difference of ORRs (sensitivity analysis).

Different equivalent margins were used according to these two different recommendations:
- Primary analysis: a two-sided 90% confidence interval (CI) for the ratio of the best ORRs at Week 24, which was calculated based on the method of logarithmic transformation. Equivalence was declared if the CI was completely within the equivalence range of (0.81, 1.24).
-Sensitivity analysis: a two-sided 95% CI for the difference of the best ORRs at Week 24 was calculated. Equivalence was declared if the CI was completely within the equivalence range of (-15%, 15%). The

clinical justification of the equivalence range is based on a regression analysis linking TTP to ORR, concluding that the (-15%, +15%) equivalence range can be translated into +/- 1.9 months TTP difference and that this difference is not considered clinically relevant.

Secondary endpoints:

- TTP (the time from randomization to date of first documentation of objective progression)
- PFS (the time from randomization to first documentation of objective progression or to death due to any cause)
- OS (the time from date of randomization to date of death due to any cause)
- DR (the time from the first documentation of objective tumour response [CR or PR] to the date of first documentation of objective tumour progression or to death due to any cause, whichever occurred first).

A sample size of **410** patients (205 per treatment group) was required to provide at least 80% power to declare MYL-1401O equivalent to Herceptin in the analysis of ORR at Week 24. This sample size assumed that both treatment groups would exhibit an ORR of 69% at Week 24 and that the ratio of MYL-1401O to Herceptin was analyzed with a two-sided 90% CI. If the 90% CI fell wholly within an equivalence region defined as (0.81, 1.24), then equivalence was to be declared.

A **formal interim analysis** was conducted when at least 30% of the information target was available (after the first 123 randomized patients enrolled under Protocol Amendment 2 [Version 5.0, 11 Oct 2013] and beyond), had either discontinued the study or completed 24 weeks in the study. The interim analyses comprised just those 123 patients.

This formal interim analysis had 2 objectives: (1) blinded sample size re-estimation (BSSR) and (2) futility analysis. The BSSR showed a best ORR of 68.3% and did not warrant sample size re-estimation. Randomization were done according to: i) tumour progression into metastatic part ≥ 2 years OR < 2 years after primary diagnosis; ii) ER/PgR status (ER- and/or PgR-positive/ER- and PgR-negative); iii) Type of taxane received (i.e., paclitaxel or docetaxel).

Disease characteristics: the majority of patients had an ECOG performance status of 0 or 1 at baseline. (Of note in the MYL-1401O group a higher percentage of patients had an ECOG of 0 compared with the Herceptin group 51.4% versus 43.7%, respectively. The majority of patients in both treatment arms was IHC positive (MYL-1401O 81.3%, Herceptin 89.0%) with a slight difference between arms. Regarding HER2/ECD status, baseline values were slightly higher in the Herceptin group compared with the MYL-1401O group (HER2/ECD ≥ 15 ng/mL: MYL-1401O 73.1%, Herceptin 78.8%). The majority of patients (80%) in both arms have received docetaxel as taxane. Tumor progression into metastatic phase and previous disease treatment were similar in both treatment arms. Tumour characteristics at baseline showed that more than half of the population had 1 or 2 metastatic sites, the majority had visceral metastasis (slightly lower in the MYL-14010 arm [74.8%] than in the Herceptin arm [81.1%]). Concomitant medications were those expected in the target population and were similar between arms.

Results:

Primary endpoint: similarity in terms of ORR at week 24 has been shown with the a priori defined margin of similarity (equivalence range of 0.81, 1.24).

Table 18 Best Overall Response Rate (ORR) at Week 24 and Ratio of Best ORR: ITT1 Population

Response		MYL-1401O + Taxane (N = 230)	Herceptin + Taxane (N = 228)
Complete response (CR)	n (%)	3 (1.3)	0 (0.0)
Partial response (PR)	n (%)	157 (68.3)	146 (64.0)
Stable disease (SD)	n (%)	48 (20.9)	49 (21.5)
Progressive disease (PD)	n (%)	9 (3.9)	20 (8.8)
N/A	n (%)	13 (5.7)	13 (5.7)
Overall response rate	n (%)	160 (69.6)	146 (64.0)
90% CI		(64.57, 74.56)	(58.81, 69.26)
95% CI		(63.62, 75.51)	(57.81, 70.26)
Ratio MYL-1401O: Herceptin		1.09	
90% CI		(0.974, 1.211)	
95% CI		(0.954, 1.237)	

CI: confidence interval, ITT: intent-to-treat, N: number of patients in treatment group, n: number of patients with data available, N/A: not applicable
Percentages are based on the number of patients in the ITT1 population.
Objective response was defined as confirmed CR or PR according to RECIST 1.1 based on central tumor evaluation. Equivalence was declared if the 90% CI of the ratio is completely within the equivalence range of (0.81, 1.24). The 90% and 95% CI was calculated on the natural log scale and back transformed using the exponential function to the original scale.

The results were considered sufficiently robust as different sensitivity analyses (including difference in ORR), supported the main analysis.

Table 19 Difference of Best Overall Response Rate (ORR) at Week 24: ITT1 Population

Response	MYL-1401O + Taxane (N = 230)	Herceptin + Taxane (N = 228)
Overall response rate, n (%)	160 (69.6)	146 (64.0)
90% CI	(64.57, 74.56)	(58.81, 69.26)
95% CI	(63.62, 75.51)	(57.81, 70.26)
Difference MYL-1401O: Herceptin	5.5	
90% CI	(-1.70, 12.69)	
95% CI	(-3.08, 14.04)	

CI: confidence interval, CR: complete response, ITT: intent-to-treat, N: number of patients in treatment group, n: number of patients with data available, PR: partial response
Percentages are based on the number of patients in the ITT1 population.
Results for difference of response rate were rounded up to one decimal point.
Objective response was defined as confirmed CR or PR according to RECIST 1.1 based on central tumor evaluation. Equivalence was declared if the 95% CI of the difference is completely within the equivalence range of (-15%, 15%).

Moreover, results of the primary analysis were further supported by secondary efficacy endpoints (TTP, PFS and OS at week 24 and week 48, as well as DR at Week 48).

TTP week 24: 35 patients (15.2%) in the MYL-1401O group had tumour progression compared to 44 patients (19.3%) in the Herceptin group. The time-to-event curves (log-rank test) for both treatment groups showed no statistically significant difference (p =0.192). For the Kaplan-Meier estimates, the median TTP was not reached due to the small number of patients who had disease progression. 95 patients (41.3%) compared with 98 patients (43.0%) had tumor progression in the MYL-1401O group and in the Herceptin group, respectively until week 48. Time-to-event curves (log rank test) for both treatment groups were not statistically significantly different (p = 0.684), and a median time to tumor progression of 11.1 months in both treatment arms was shown by Kaplan Meier estimates.

Subgroup analyses were supportive of the overall population results. Some subgroups seemed to favour MYL-1401O but in view of limited numbers, no conclusion could be made. Sensitivity analyses using different population sets showed consistent results with the primary analysis.

PFS week 24: 189 patients (82.2%) compared with 180 patients (78.9%) had PFS in the MYL-1401O group and in the Herceptin group, respectively. Time-to-event curves for both treatment groups were not statistically significantly different (p = 0.303). The median time for PFS (Kaplan Meier estimates) was not reached due to the limited number of patients who had tumour progression.
Until Week 48, 128 patients (55.7%) and 126 patients (55.3%) in the MYL-1401O group and in the Herceptin group, respectively, did not have progression of the disease. Time to-event curves for both treatment groups were not statistically significantly different (p = 0.842) and the median time for PFS (Kaplan-Meier estimates) was 11.1 months in both treatment arms.
Subgroups analyses supported the results from the overall population. The same subgroups seen for the primary analysis, i.e., previous adjuvant/neoadjuvant chemotherapy and tumour endocrine status negative seemed to favour MYL-1401O, but no clinical significance could be established in view of the limited number of events.

Updated analysis of PFS showed that at the final assessment, 82 patients (35.7%) in the MYL-1401O group were free of progression compared with 86 patients (37.7%) in the Herceptin group. The time-to-event curves for both treatment groups were not statistically significantly different (p = 0.864, log-rank test). The median time for PFS (Kaplan-Meier estimates) was 11.1 months in both groups. The hazard ratios (unstratified and stratified) MYL-1401O: Herceptin (Cox proportional hazards model) were not statistically significant.

OS: Until Week 48, 205 patients (89.1%) survived in the MYL-1401O group compared with 194 patients (85.1%) in the Herceptin group (the survival curves for both treatment groups were not statistically significantly different p = 0.131).
For the Kaplan-Meier estimates for OS, the median was not reached due to the limited number of patients in the ITT1 population who died prior to analysis.

At final data cut, 121 patients (52.6%) survived in the MYL-1401O arm compared to 114 patients (50.0%) in the Herceptin arm. The log-rank test did not show statistically significant differences in both arms (p=0.427) of the study. Median time for OS (Kaplan-Meier estimates) was 35.0 months in the MYL-1401O arm compared to 30.2 months in the Herceptin. The hazard ratio was 0.90 (unstratified) and 0.87 (stratified) at the time of final OS analysis. Thus, OS was slightly longer for MYL-1401O compared with Herceptin, although not statistically significant.

Duration of response: 81 patients (42.4%) the MYL-1401O group and 81 patients (44.5%) in the Herceptin group with objective response had tumor progression at week 48 or died before the 48th week cut-off. According to the log-rank test, the time-to-event curves for both treatment groups were not statistically significantly different (p = 0.790).
Median time (Kaplan-Meier estimates) to tumor progression or death after objective tumor response was 9.7 months in both treatment arms (more than 50% of patients in both treatment arms did not have tumor progression or did not die until Week 48). The updated analysis at the end of study showed that 123 patients (64.1%) and 119 patients (64.7%) in the MYL-1401O and in the Herceptin group, respectively, with objective response had tumor progression or died. According to the log-rank test, the time-to-event curves for both treatment groups were not statistically significantly different (p = 0.771). Median time (Kaplan-Meier estimates) to tumor progression or death after objective tumor response was 9.9 months in the MYL-1401O group and 9.8 months in the Herceptin group. The hazard ratios MYL-1401O: Herceptin obtained from the Cox proportional hazard model were 0.96 (unstratified) and 1.00 (stratified) at the time of the final analysis with p-values > 0.05 for both hazard ratios. Thus, the average hazard rate for tumor progression or death after the tumor response was not statistically significant.

Conclusion on the clinical efficacy

Similarity in terms of ORR at week 24 was shown with the a priori defined margin of similarity (equivalence range of 0.81, 1.24) being met. The results were considered robust enough as different sensitivity analyses supported the main analysis. Moreover, results of the primary analysis were further supported by the secondary efficacy endpoints.

Overall, the clinical efficacy data supported biosimilarity.

Clinical safety

The dossier comprises three clinical trials, MYL-Her-1001 MYL-Her-1002 MYL-Her-3001 and a supportive study Bmab M200-CT3-001-11. SRA-approved reference products (EU-approved Herceptin and/or US-licensed Herceptin) were used as the reference products in these studies.

The safety evaluation include data from all subjects who received at least one dose of the study drug.

Safety data from these studies were not pooled for analysis, because the studies were too heterogeneous in terms of population, treatment duration, and study drug formulation.

Studies MYL-Her-1001 and MYL-Her-1002 were conducted in healthy male volunteers, enrolling 22 and 132 subjects, respectively. A single dose of 8 mg/kg by intravenous infusion of Biocon's trastuzumab (MYL-1401O) or Herceptin was administered. These studies were designed to determine the PK/PD of Biocon's Trastuzumab (MYL-1401O).

The main safety data emerged from two in-patient studies. Study MYL-Her-3001 which enrolled 500 patients with HER2-positive metastatic breast cancer and the supportive study BM200-CT3-001-11 which enrolled 134 patients with HER2-positive metastatic breast cancer. Patients received a loading dose of 8 mg/kg by IV infusion, followed by maintenance dose of 6 mg/kg, every 3 weeks.

Study BM200-CT3-001-11 used a different formulation than study MYL-Her-3001. Bmab 200 was the initially developed product used in clinical study BM200-CT3-001-11 to obtain marketing authorization in India. That formulation was developed using same excipients as in the reference product formulations.

The safety parameters evaluated included: adverse events (AEs); haematology; biochemistry serum values; urinalysis; vital signs; Draize scale for local tolerance at intravenous injection site; C-reactive protein (CRP); immunoglobulins (IgA, IgM, IgG); echocardiography (left ventricular ejection fraction [LVEF]); and immunogenicity (anti-drug antibodies [ADA] against trastuzumab)

Patient exposure was balanced in both arms of the studies conducted in healthy volunteers.

In Study MYL-Her-3001, the mean and medium exposure was higher in the Biocon's trastuzumab arm than in the reference product arm. In Study BM200 CT3-001-11, exposure appears similar in both arms.

In study MYL-Her-3001, the proportion of patients with Grade ≥ 3 treatment-emergent adverse events (TEAEs) and with serious TEAEs was comparable between the two treatment arms; however, the proportion of patients with treatment-related TEAEs was higher in the Biocon's trastuzumab arm (41.7%) compared with the reference product arm (35.8%)

In study BM200-CT3-001-1, the most common TEAEs in both arms were pyrexia (18.18% for the Biocon's trastuzumab arm, 22.06% for the Herceptin arm) and diarrhoea (16.67% for the Biocon's trastuzumab arm and 14.71% for the Herceptin arm).

Overall, there were some numerical differences in the incidence of TEAEs between treatment arms: headache, anaemia and pyrexia being higher in the Biocon's trastuzumab arm than the Herceptin arm; infections and infestations were lower in the Biocon's trastuzumab. The reported TEAEs were in line with the known safety profile of trastuzumab.

Infusion-related reactions, pulmonary toxicity and cardiac toxicity were similar between treatment arms. The incidence of pulmonary, cardiac toxicity was higher in subjects who had previously received anthracycline therapy and in subjects with pre-existing conditions (e.g. had a previous or concomitant cardiovascular disorder, previous thoracic radiation, diabetes mellitus, or high levels of blood pressure) and/or disease progression.

The incidence of SAEs was similar between treatment arms, 97 patients (39.3%) in the Biocon's trastuzumab (MYL-1401O) arm and 91 patients (37.0%) in the reference group. Most SAEs that began in Part 1 resolved or resolved with sequelae, except for those that were fatal.

In study MYL-Her-3001, for Part 1 and 2 through Week 48, 10 patients experienced fatal TEAEs, 6 in the MYL-1401O arm (2.4%, 8 events) and 4 in the Herceptin arm (1.6%, 6 events). Most deaths were attributed to concomitant medications, underlying conditions or disease progression. Three respiratory failure events were reported in study MYL-Her-3001. Two events respiratory failure (one in each arm) were considered probably related to the study drug.

In Study BM200-CT3-001-11, 4 deaths were reported. Two deaths occurred in the Bmab-200 arm: disease progression and multi-organ failure, which were considered not related to the study drug. Two deaths also occurred in the Herceptin arm: disseminated intravascular coagulation, which was considered to be possibly related to the study drug, and sepsis which was considered not related to the study drug.

There were no clinically significant changes in haematology, biochemistry and urinalysis throughout the clinical development program of Biocon's trastuzumab. With respect to blood and lymphatic system

disorders, neutropenia was reported most frequently (56.0%), being Grade 1 and 2 in intensity and considered unrelated to the study drug.

In summary, no new unexpected AEs occurred in the Bmab-200 arm, and the relative incidence of different types of AEs is similar to Herceptin, in this supportive study.

Immunogenicity was evaluated as per the EMA guideline on *Immunogenicity assessment of biotechnology-derived therapeutic proteins* (EMEA/CHMP/BMWP/14327/2006 Rev)

Overall, anti-drug antibody titers were low, transient and similar between the Biocon's trastuzumab (MYL-1401O) and reference product. This is consistent with the known immunogenicity potential of trastuzumab.

The incidence of TEAEs leading to study drug discontinuation was slightly higher in the Herceptin arm (12 patients, 7.3%) than in the Biocon's trastuzumab (MYL-1401O) arm (7 patients, 3.9%) in study MYL-Her-3001. The incidence of TEAEs leading to study drug discontinuation was low in subjects receiving monotherapy.

The events leading to discontinuation included: infusion related reactions, and cardiac and pulmonary toxicities.

Conclusion on clinical safety

Biocon's trastuzumab demonstrated a safety profile similar to Herceptin.

Overall conclusion on clinical data

The clinical part is accepted.

6. Risk Management Plan (RMP)

The submitted RMP complies with EMA *Guidance on the format of the risk management plan*, and is acceptable. The RMP also includes a WHO PQ-specific addendum and this is considered acceptable.

7. Pharmacovigilance plan

There are no planned or ongoing additional studies included in the pharmacovigilance plan; only routine pharmacovigilance activities are proposed by the applicant. This is in line with the reference product pharmacovigilance plan and is acceptable.

8. Benefit Risk Assessment

Based on WHO's assessment of quality, non-clinical and clinical data, the team of assessors considered that the benefit–risk profile of [BT-ON013 trade name] was acceptable for the following indication: **'treatment of early stage HER2 positive breast cancer or metastatic HER2 positive breast cancer'**, and advised that [BT-ON013 trade name] is highly similar to the reference product Herceptin with respect to quality, safety and efficacy. As such, [BT-ON013 trade name], manufactured at Biocon Limited, Special Economic Zone, Plot Nos. 2, 3, 4 & 5, Phase-IV, Bommasandra -Jigani Link Road,Bommasandra Post, Bengaluru – 560 099, India, has been included in the list of prequalified medicinal products.