International Journal of BioAnalytical Methods & BioEquivalence Studies (IJBMBS)
ISSN 2470-4490

Bioequivalence Study of Dexrabeprazole Gastro Resistant Tablets in Healthy Male Subjects Under Fasting and Fed Conditions

Research Article

G. Demiray[1*], P. Özdüven[2], E. Durucu[3], M. Bilgiç[4], H. Aydonat[1], B. Güney[1], E. Doğan-Kurtoğlu[1], S. Bilgiç[4], R.S. Alpan[2], M. Nacak[3], O. Sağlam[1], A. Erenmemişoğlu[5]

[1] Novagenix Bioanalytical Drug R&D Centre, Ankara, Turkey.
[2] TNC Pharmaceuticals R&D Ltd. Istanbul, Turkey.
[3] Farmagen-Good Clinical Practice Center, Gaziantep, Turkey.
[4] Neutec İlaç San. Tic. A.Ş.-Turkey.
[5] Alpan Farma R&D Biotechnologies Ltd., Istanbul, Turkey.

Abstract

Rabeprazole is a racemic mixture of two enantiomers, R(+)-enantiomer and S(-)-enantiomer. The active ingredient of the test product formulation is dexrabeprazole, the chirally pure R(+)-enantiomer of rabeprazole. This study aims to compare the pharmacokinetic properties ofdexrabeprazole containing product (i.e. test product) to racemate rabeprazole containing product (i.e. reference product) to demonstrate the bioequivalence with respect to the rate and extent of absorption of dexrabeprazolein healthy male subjects under fasting and fed conditions. Under fasting conditions; for test and reference products, the mean±sd of C_{max} were found 288.374±79.3044 ng/mL and 288.255±89.8262 ng/mL, and the mean±sd of $AUC_{(0\text{-tlast})}$ were found 504.308±211.2707 hr.ng/mL and 572.973±246.9999 hr.ng/mL, respectively. Under fed conditions; for test and reference products, the mean±sd of C_{max} were found 301.094±136.685 ng/mL and 304.202±134.168 ng/mL, and the mean±sd of AUC(0-tlast) were found 576.533± 269.297 hr.ng/mL and 660.652±298.974 hr.ng/mL, respectively. The primary target variables data demonstrate the bioequivalence of test and reference products with regard to 90% CI for C_{max} of 95.23-107.79 and for $AUC_{(0\text{-tlast})}$ of 84.83-90.41 under fasting conditions and for C_{max} of 88.23 – 109.98 and for $AUC_{(0\text{-tlast})}$ of 81.55 – 96.42 under fed conditions. It was shown that the test product containing dexrabeprazole alone (Rabby-D 10 mg enteric coated tablet, NeutecİlaçSan. Tic. A.Ş.-Turkey) and reference product containing racemate rabeprazole (Pariet® 20 mg gastro-resistant tablet, Eisai Limited European Knowledge Centre-UK) are bioequivalent in terms of rate and extent of absorption for dexrabeprazole under fasting and fed conditions. Besides, both products were well tolerated and safe.

Keywords: Deksrabeprazole; Rabeprazole; Bioequivalence; Gastro-Resistant Tablet; Method Validation.

Introduction

Rabeprazole sodium is a proton pump inhibitör, which is a substituted benzimidazole known chemically as 2-[[[4-(3methoxypropoxy)-3-methyl-2-pyridinyl]-methyl]sulfinyl]-1H–benzimidazole sodium salt. Rabeprazole is a racemic mixture of two enantiomers, R(+)-enantiomer and S(-)-enantiomer. The active ingredient of the test product formulation is dexrabeprazole, the chirally pure R(+)-enantiomer of rabeprazole, which was shown more effective than the racemate and S(-)-rabeprazole ininhibiting acid-related gastric lesions in rats [1]. Recent studies have revealed that the pharmacodynamics of (R)-(+)-rabeprazole were better than those of (S)-(−)-rabeprazole [1-4].

Absorption of rabeprazole is rapid, with peak plasma levels occurring approximately 3.5 hours after a 20 mg dose. Peak plasma concentrations (C_{max}) of rabeprazole and AUC are linear over the dose range of 10 mg to 40 mg. Absolute bioavailability of an oral 20 mg dose (compared to intravenous administration) is about 52% due in large part to pre-systemic metabolism. In healthy subjects the plasma half-life of rabeprazole is approximately one hour (range 0.7 to 1.5 hours), and the total body clearance is estimated to be 283 ± 98 ml/min. Neither food nor the time of

***Corresponding Author**:
Gökçe Demiray,
Novagenix Bioanalytical Drug R&D Centre, Ankara, Turkey.
E-mail: gdemiray@novagenix.com

Received: November 04, 2022
Accepted: February 09, 2023
Published: February 20, 2023

Citation: G. Demiray, P. Özdüven, E. Durucu, M. Bilgiç, H. Aydonat, B. Güney, et al., Bioequivalence Study of Dexrabeprazole Gastro Resistant Tablets in Healthy Male Subjects Under Fasting and Fed Conditions. *Int J Bioanal Methods Bioequival Stud.* 2023;6(1):93-99.

day of administration of the treatment affect the absorption of rabeprazole sodium.

Rabeprazole sodium, as is the case with other members of the Proton pump inhibitors (PPI class of compounds), is metabolised through the cytochrome P450 (CYP450) hepatic drug metabolising system. In vitro studies with human liver microsomes indicated that rabeprazole sodium is metabolised by isoenzymes of CYP450 (CYP2C19 and CYP3A4). In these studies, at expected human plasma concentrations rabeprazole neither induces nor inhibits CYP3A4. In humans the thioether (M1) and carboxylic acid (M6) are the main plasma metabolites with the sulphone (M2), desmethyl-thioether (M4) and mercapturic acid conjugate (M5) minor metabolites observed at lower levels. Only the desmethyl metabolite (M3) has a small amount of anti-secretory activity, but it is not present in plasma. Following a single 20 mg 14C labelled oral dose of rabeprazole sodium, no unchanged drug was excreted in the urine. Approximately 90% of the dose was eliminated in urine mainly as the two metabolites: a mercapturic acid conjugate (M5) and a carboxylic acid (M6), plus two unknown metabolites. The remainder of the dose was recovered in faeces [5].

Clinicalbioequivalence studiesarerequired for dexrabeprazole containing orally administered equivalent products according to local and globalpharmaceutical regulations [6, 7]. Therefore, two studies under fasting and fed conditions were conducted to demonstrate the bioequivalence of the products with respect to the rate and extent of absorption of deksrabeprazole in healthy male subjects under fasting and fed conditions.

Subjects and Methods

Ethical Statement

This study was conducted at Farmagen-Good Clinical Practice Center, Gaziantep, Turkey according to the regulations by Ministry of Health of the Republic of Turkey which are in compliance with Declaration of Helsinki and Good Clinical Principles (GCP) [8]. The protocols and informed consent forms werereviewed and approved byan independent ethics committee (Erciyes University, Bioavailability-Bioequivalence Research Ethics Commitee, Kayseri, Turkey,Approval Date: 17.02.2021for fasting and 09.06.2021 for fed conditions) and Turkish Medicines and MedicalDevices Agency (Approval Date: 06.04.2021 for fasting and 17.06.2021 for fed conditions). All subjects voluntarily provided signed informed consent before participation in thestudy.

Study Population and Study Design

All subjects are adult males (aged 18-55 years) with normal weight according to the a body mass index BMI. The subjects who have history of drug hypersensitivity (especially to the active and inactive ingredients of the rabeprazole sodium preparations or intolerance to any sugar) and who have any history or presence of clinically significant cardiovascular, renal, hepatic, pulmonary, metabolic, endocrine, hematological, gastrointestinal, neurological, psychiatric or other diseases were excluded from the study. The inclusion and exclusion criteria were established clearly together with the reasons for withdrawal from the study. The subjects who were willing to participate in the clinical trial signed the informed consent form on their own freewill and understood that they could withdraw from the study anytime without specifying any reason. Two of the studies were conducted asamonocentric, open-label, randomised, single oral dose, four-period replicate, crossover, study in 36 healthy, Caucasian, adult, male, human subjects under fasting and fed conditions. Both studies consisted of 7 days including 2-days isolation and four consecutive study periods with a hospitalization of approximately 115 hours and wash-out between periods "a-day". Studies were conducted at Farmagen-Good Clinical Practice Center, Gaziantep, Turkey. The standard laboratory examinations in blood and urine were done consistent with the study protocol and the volunteers were checked for presence of HBsAg, HCV-Ab and HIV-Ab in serum. Also Covid-19 PCR tests were applied to the volunteers before isolation period and hospitalization. Volunteers were also requested to provide a urinesample for a drug screen which include "amphetamines, cannabinoids, benzodiazepines, cocaine, opioids and barbiturates" and an alcohol breath test before isolation periods. All laboratory tests were carried out in a certified local laboratory. A total of 36 subjects in each study have been randomised.

Volunteers were isolated for two days at the dorm/hotel; observed for their well-beings confirming with vital sign measurements; checking for exclusion criteria and registration of any concomitant medication. During the isolation period volunteers consumed the meals served by dorm/hotel administration comply with the restricted foods and beverages defined in the protocol. After two night isolation period volunteers were discharged from dorm/hotel and transferred to the clinic for hospitalization. A confirmation swab test for COVID-19 before hospitalisation was assessed. Volunteers were admitted to the clinic with negative PCR test result.

An evening meal was provided at hospitalization days (total caloric value of approximately 1200 kcal). On medication days, a standard lunch (total caloric value is approximately 1200 kcal) was provided 4 hours after dosing, and a standard evening meal (total caloric value is approximately 1200 kcal) was provided 10 hours after dosing in each period. In the study which conducted underfed conditions a high-fat, high-caloric breakfast (total of approximately 900 to 1000 kcal) was provided between 7:30 and 8:30 a.m. on Day 1 of each period before administration of study medications.

Investigational Medicinal Products

The test drug used was Rabby-D 10 mg enteric coated tablet (gastroresistant tablet), NeutecİIaç San. Tic. A.Ş.-Turkey (Batch No: NI00190-2104 P02; Expiry Date:04.2023); the reference drug used was Pariet® 20 mg gastro-resistant tablet, Eisai Limited European Knowledge Centre-UK (Batch No: 127525; Expiry Date: 03.2022).

Blood sampling and Study Assessment

The samples were drawn by a short intravenous catheter at predose and 1:00, 1:30, 2:00, 2:20, 2:40, 3:00, 3:20, 3:40, 4:00, 4:30, 5:00, 6:00, 7:00, 8:00, 9:00, 10:00, 11:00, 12:00 and 14:00 hours post-dose in each clinical study period for fasting conditions and at predose and 1:00, 2:00, 3:00, 3:30, 4:00, 4:30, 5:00, 5:30, 6:00, 6:30, 7:00, 7:30, 8:00, 8:30, 9:00, 10:00, 11:00, 12:00, 14:00 and 16:00 hours post-dose in each clinical study period for fed conditions. The blood samples (5 ml) were collected into tubes con-

taining K2 EDTA as anti-coagulating agent. After sampling, the samples were immediately refrigerated at approximately 2-8°C and will remain there for not more than 20 minutes. Following the centrifugation (1500 g, 4°C, 10 min), the separated plasma from each sample weretransferred into two 3.5 mL transparent, polypropylene tubes. All thealiquoted plasma samples were flash freezed immediately. The flash frozen samples (aliquoted plasma samples) were transferred to a deep-freezer and stored at -70°C until they were transported to the bioanalytical center.

Determination of deksrabeprazole plasma concentrations

The bioanalytical phase of the study has been run at Novagenix Bioanalytical R&D Center, Ankara, Turkey. In order to avoid bias, the analytical studies were operated as analytically blinded.

The method used for the determination of dexrabeprazole was developed and validated by Novagenix Bioanalytical R&D Center, Ankara in accordance with the earlier published method on stereoselective pharmacokinetics of rabeprazole [9].

The lower limit of quantification for dexrabeprazole was 1 ng/mL. Standard curve range for dexrabeprazole was 1 ng/ml to 1200 ng/ml. Dexrabeprazole was extracted from plasma by protein precipitation using acetonitrile. Finally, the samples were transferred into a collection plate for analysis.

Materials and Methods

Chemicals and reagents

Reference standard "Dexrabeprazole sodium"was supplied from Nosch Labs Private Limited, India (Lot no:DRS0050617 with a retest date of 1st of June 2022) and internal standard (R)-Rabeprazole-d3 Sodium Salt (Cas no:1216494-11-9, certified purity 95%) was obtained from Toronto Research Chemicals, Canada. Methanol Acetonitrile ammonium acetate and acetic acid were purchased from Merck KGaA, Darmstadt, Germany. Ultrapure (Type 1) water was obtained from Milli-Q plus water purification system. K2EDTA blank human plasma including haemolysed and hyperlipidaemic were purchased from Bioivt Elevating Science,UK and Gaziantep University Farmagen GCP Centre, Turkey.

Instrument and Conditions

Analyses were performed on a LC-MS/MS system consisting of a Shimadzu mass spectrometer LCMS-8050 coupled to triple quadrupole mass spectrometry detector with an electrospray ionization (ESI) interface and NexeraX2 model LC system (SIL-30AC autosampler, LC-30AD solvent delivery modules, CTO-10AS vp column oven, DGU-20A5R degasser unit; Shimadzu Corporation. Japan). All data were processed by Shimadzu LabSolution Software version 5.93 (Shimadzu Corporation. Japan).

Chromatographic separations were achieved by using a Daicel ChiralPAK IC, 4.6 mm I.D. x 150 mmL, 5 µm column and mobile phase was consisting of acetonitrile and 0.2% acetic acid in 10 mM ammonium acetate solution (80:20, v:v). Flow rate was 0.8 mL/min and the chromatographic run time was 6.5 minutes. Column temperature was set to 30°C and the autosampler temperature was 10°C.

The determination of dexrabeprazole is performed in tandem mass spectrometry operated in the positive ion electrospray ionisation (ES+).The multiple reaction monitoring (MRM) transitions were performed at m/z359.90>242.20 for dexrabeprazole and m/z363.00>245.20 for dexrabeprazole d3.

Preparation of Standard and Quality Control (QC) samples

Stock solutions of (R)-rabeprazole sodium salt were prepared in methanol:water (1:1) mixture separately for calibration standards and quality control samples. Final concentrations were 1 mg/mL and diluted working stock solutions were prepared in methanol. Internal standard (IS) stock solution was prepared by dissolving (R)-rabeprazole d3 sodium salt in methanol:water (1:1) mixture and the final concentration was 0.2 mg/mL stock solution.

Calibration standards were prepared for the concentration levels of 1, 2, 20, 100, 250, 500, 1080 and 1200 ng/mL and quality control samples were prepared for the concentration levels of 1, 3, 30, 480 and 960 ng/mL.

Sample Preparation

100 µL plasma was spiked with 50 µL IS working solution (0.7 µg/mL) then protein precipitation was applied with 600 µL acetonitrile. After centrifugation for 10 minutes at 5500 rpm (4°C), 2 µL upper organic phase was injected to the system.

Method Validation

The method was completely validated according to US-FDA Bioanalytical Method Validation Guidance [10] and European Medicines Agency Guideline on Bioanalytical Method Validation [11]. The parameters (selectivity, linearity, lower limit of quantification, accuracy, precision, dilution integrity, influence of haemolysed and hyperlipidaemic plasma, drug-drug interaction, carry-over, recovery, matrix effect,re-injection reproducibility, batch size, stability of the analyte) were successfully validated.

For selectivity, eight different sources of human blank plasma (including haemolysed and hyperlipidaemic) were evaluated and no interference was observed at the retention times and transitions of dexrabeprazole and dexrabeprazole d3. Eight freshly prepared calibration standards for dexrabeprazole (1, 2, 20, 100, 250, 500, 1080, 1200 ng/mL) were assayed in each of three validation batches. For each validation batch, a calibration curve was acquired by plotting the peak area ratios (peak area analyte/peak area IS) versus nominal concentration and fitted into the linear equation using weighing factor 1/C2 as the best fit model for this curve. The range of precision and accuracy of the back-calculated concentrations of the calibration curve points were from 0.52% to 1.84% and from 95.76% to 105.40%, respectively.

The within-batch precision and accuracy were evaluated by analyzing QC samples at five different concentration levels with six replicates in a batch.The between-batch precision and accuracy were determined by analyzing three different batches. The within-batch accuracy and precision was 96.70% to 106.94% and 0.45% to 5.99%, respectively. The between-batch accuracy and precision was 97.44% to 105.31% and 0.84% to 3.56%, respectively.

Stability evaluation in matrix were processed using freshly prepared calibration standards and freshly prepared QC samples. Dexrabeprazole was stable in plasma at room temperature for 5 hours and after 4 freeze thaw cycles. The processed samples were stable up to 30 hours in autosampler at 10 °C. Dexrabeprazole was stable in plasma for at least 92 days when stored at -20 ºC and -70 ºC.

Pharmacokinetic and statistical analyses

In accordance with the bioequivalence recommendation on rabeprazole sodium delayed-release tablets and the earlier published assessment reports for rabeprazole sodium containing generic products, the intra-subject coefficient of variation (ISCV) was estimated higher than 50% for C_{max} and approximately 30% for $AUC_{(0\text{-tlast})}$. In order to demonstrate bioequivalence with a power of 80% and a test/reference parameter ratio as 0.95 for a fullyreplicated crossover design, sample size was estimated as 30. Considering the possible drop-outs, a sample size of '36 volunteers' was chosen in a replicate design.

Maximum plasma concentration (C_{max}) and area under the curve from time 0 to the last measurable concentration ($AUC_{(0\text{-tlast})}$) were considered as the primary target variables; area under the curve from time 0 to the infinite time ($AUC_{(0\text{-}\infty)}$), time to reach the peak concentration (t_{max}), terminal half life ($t^{1}/_{2}$), terminal disposition rate constant (λz) and mean residence time (MRT) were declared as the secondary target variables in this bioequivalence study.

C_{max} and tmax for dexrabeprazole were obtained directly by plasma concentration-time curves. $AUC_{(0\text{-tlast})}$ was calculated using the linear-log trapezoidal rule. $AUC_{(0\text{-}\infty)}$ was calculated by summing $AUC_{(0\text{-tlast})}$ and extrapolated area. The latter was determined by dividing the last measuredconcentration by λz which was estimated byregression of the terminal log-linear plasma concentration time points.

Databases were automatically processed using the validated program of Phoenix WinNonlin® version8.3.1.5014 (Certara Inc., Pharsight,USA). Obtained PK parameters were processed using the SAS® software version 9.0.

An analysis of variance (ANOVA) was performed using the General Linear Model (GLM) procedure, in which sequence, subject (nested in sequence), period and treatment effects were characterized. The effects of ANOVA were tested at 5% level of significance. The 90% confidence intervals were calculated for T/R ratio of means. The 90% confidence interval of geometric least square means ratio T/R for $AUC_{(0\text{-tlast})}$ and C_{max} for dexrabeprazole were the primary parameters for the bioequivalence assessment.

In the assessment of bioequivalence, confidence intervals approach was used. The two one-sided hypothesis at the 5% level of significance were tested by constructing the 90% confidence intervals (90% CIs) for the geometric mean ratios of test/reference products. , In addition, a non-parametric Wilcoxon and median tests for tmaxwereperformed using SAS procedure NPAR1WAY. Both databases were automatically processed using the validated program of Phoenix WinNonlin® version 8.3.1.5014 (Certara Inc., Pharsight, USA). Obtained PK parameters were processed using the SAS® software version 9.0

Results

For fasting study, 51 subjects were screened. 36 subjects were randomised and included into the study. The subjects were divided into two groups according to the randomisation table. There was no drop-out and 36 subjects completed the clinical phase of the study. All of the subjects were Caucasian. The mean±SD age of subjects was 23.72±6.44 years and the mean±SD body mass index (BMI) was 24.04±3.15.

For fed study, 47 subjects were screened. 36 subjects were randomised and included into the study. The subjects were divided into two groups according to the randomisation table. There was two drop-outs and 34 subjects completed the clinical phase of the study. All of the subjects were Caucasian. The mean±SD age of completed subjects was 26.9±8.50 years and the mean±SD BMI was 24.7±3.15.

The demographic data of subjects are presented in Table 1. There was no protocol deviation through the clinical period.

Table 1. Demographic data of the subjects.

UNDER FASTINGCONDITIONS (n=36)				
	Age(year)	**Weight(kg)**	**Height(cm)**	**Body mass index**
Mean	23.72	73.81	175.19	24.04
SD	6.44	10.41	4.67	3.15
Minimum	18	60	164	19.2
Maximum	43	98	182	29.7
UNDER FED CONDITIONS (n = 34)				
	Age(year)	**Weight(kg)**	**Height (cm)**	**Body mass index**
Mean	26.9	74.2	173.2	24.7
SD	8.5	11	6.6	3.15
Minimum	18	51	162	18.7
Maximum	49	93	185	29.8

Actual time of sampling was used in the estimation of the pharmacokinetic parameters.

In period 2, 3 and 4 at t0.00, the concentrations were found as zero or <LLOQ or less than 5% C_{max} for all of the subjects indicating the absence of carry over effect and the washout period of 1 day was sufficient under fasting and fed conditions.

The pharmacokinetic parameters for test and reference products are summarised in Table 2 and Table 3 for fasting and fed conditions, respectively. The geometric least square means, ratios and 90% CIs are summarised in Table 4 and Table 5 for fasting and fed conditions, respectively.

Average plasma concentration-time curves and average ln plasma concentration-time curves of test and reference products for single dose of dexrabeprazole under fasting conditions are displayed in Figure 1, respectively.

Average plasma concentration-time curves and average ln plasma concentration-time curves of test and reference products for single dose of dexrabeprazole under fed conditions are displayed in Figure 2, respectively.

Under fasting conditions; for Test and Reference products, the mean±sd of Cmax were found 288.374±79.3044 ng/mL and 288.255±89.8262 ng/mL, and the mean±sd of $AUC_{(0\text{-tlast})}$ were found 504.308±211.2707 hr.ng/mL and 572.973±246.9999 hr.ng/mL, respectively (Table 2).

The ln-transformed geometric least square means ratio (test/reference, point estimator) for $AUC_{(0\text{-t})}$ was 87.57% and the 90% confidence interval was 84.83% to 90.41%. The ln-transformed geometric least square means ratio (test/reference, point estimator) for C_{max} was 101.31% and the 90% confidence interval was 95.23% to 107.79%. Thus, the confidence intervals for $AUC_{(0\text{-t})}$ and C_{max} ratios were within the standard acceptance range of 80.00 – 125.00%[4] (Table 4).

Table 2. The pharmacokinetic parameters of dexrabeprazole after oral administrations of Test Product containing 10 mg dexrabeprazole sodium and Reference Product containing 20 mg rabeprazole sodium in healthy male subjects under fasting conditions (N=36, 72 observations).

Test Product				
(Rabby-D 10 mg enteric coated tablet (gastro-resistant tablet), NeutecİIaç San. Tic. A.Ş., Turkey)				
Parameters (Units)	**Geometric Mean**	**Arithmetic Mean±SD**	**Range**	**Median**
C_{max}(ng/mL)	277.57	288.374±79.304	139.965 - 513.042	280.849
$AUC_{(0\text{-tlast})}$(ng.hr/mL)	466.7	504.308±211.271	176.414 - 1239.154	478.376
t_{max} (hr)	2.346	2.461±0.835	1.000 - 6.000	2.333
$t_{1/2}$ (hr)	1.711	1.910 ± 0.901	0.581 - 5.100	1.859
Reference Product				
(Pariet® 20 mg gastroresistant tablet, Eisai Limited European Knowledge Centre, United Kingdom)				
C_{max}(ng/mL)	273.976	288.255±89.826	91.279 - 558.073	281.208
$AUC_{(0\text{-tlast})}$(ng.hr/mL)	532.92	572.973±246.999	268.647 - 1566.714	531.695
t_{max} (hr)	3.513	3.611±0.872	2.000 - 6.000	3.333
$t_{1/2}$ (hr)	2.023	2.181 ± 0.821	0.717 - 4.424	2.133

Table 3. The pharmacokinetic parameters of dexrabeprazole after oral administrations of Test Product containing 10 mg dexrabeprazole sodium and Reference Product containing 20 mg rabeprazole sodium in healthy male subjects under fed conditions (N=34, 68 observations).

Test Product				
(Rabby-D 10 mg enteric coated tablet (gastro-resistant tablet), NeutecİIaç San. Tic. A.Ş., Turkey)				
Parameters (Units)	**Geometric Mean**	**Arithmetic Mean±SD**	**Range**	**Median**
C_{max}(ng/mL)	284.586	301.094±136.685	0.000- 811.190	304.17
$AUC_{(0\text{-tlast})}$(ng.hr/mL)	514.729	576.533±269.297	114.707 - 1418.870	544.773
t_{max} (hr)	4.208	4.662±2.817	0.000 - 16.000	4
$t_{1/2}$ (hr)	1.694	1.907 ± 0.962	0.653- 4.945	1.583
ReferenceProduct				
(Pariet® 20 mg gastroresistant tablet, Eisai Limited European Knowledge Centre, United Kingdom)				
C_{max}(ng/mL)	292.2	304.202±134.168	0.000- 655.729	305.007
$AUC_{(0\text{-tlast})}$(ng.hr/mL)	593.11	660.652±298.974	97.040- 1806.212	595.992
t_{max} (hr)	4.1	4.493±2.841	0.000 - 16.000	4
$t_{1/2}$ (hr)	1.879	2.120 ± 1.125	0.701 - 5.829	1.851

For the secondary endpoint data, the median of t_{max} for Test and Reference product were found 2.333 hr and 3.333 hr, respectively and ranged from 1.00 hr to 6.00 hr and from 2.00 hr to 6.00 hr, respectively. Besides, the mean±sd of t1/2 for Test and Reference product were found 1.910 ± 0.901 hr (ranged from 0.581 hr to 5.100 hr) and 2.181 ± 0.821 hr (ranged from 0.717 hr to 4.424 hr), respectively (Table 2).

Under fed conditions; for Test and Reference products, the mean±sd of Cmax were found 301.094±136.685 ng/mL and 304.202±134.168 ng/mL, and the mean±sd of $AUC_{(0\text{-}tlast)}$ were found 576.533± 269.297 hr.ng/mL and 660.652±298.974 hr.ng/mL, respectively (Table 3).

The ln-transformed geometric least square means ratio (test/reference, point estimator) for $AUC_{(0\text{-}t)}$ was 88.68% and the 90% confidence interval was 81.55% to 96.42% %. The ln-transformed geometric least square means ratio (test/reference, point estimator) for C_{max} was 98.50% and the 90% confidence interval was 88.23% to 109.98%. Thus, the confidence intervals for $AUC_{(0\text{-}t)}$ and C_{max} ratios were within the standard acceptance range of 80.00 – 125.00%. (Table 5).

For the secondary endpoint data, the median of t_{max} for both Test and Reference product were found 4.000 hr and ranged from 0.00 hr to 16.00 hr. Besides, the mean±sd of t1/2 for Test and Reference product were found 1.907 ± 0.962 hr (ranged from 0.653 hr to 4.945 hr) and 2.120 ± 1.125 hr (ranged from 0.701 hr to 5.829 hr), respectively (Table 3).

Statistical Parameters

Tables and Figures 1 & 2

Discussion

For fasting conditions; the results show that intra-individual vari-

Table 4. Geometric Least Square Means point estimator, 90% Confidence Intervals, %CVintra and Powerfor primary endpoints of fasting conditions.

Endpoint	Point estimator (%)	90% Confidence Interval	%CVintra	Power (%)
Cmax(ratio test/reference)	101.31	95.23-107.79	22.68	>99.99
AUC(0-tlast)(ratio test/reference)	87.57	84.83-90.41	11.55	>99.99

Table 5. Geometric Least Square Means point estimator, 90% Confidence Intervals, %CVintra and Powerfor primary endpoints of fed conditions.

Endpoint	Point estimator (%)	90% Confidence Interval	%CVintra	Power (%)
Cmax(ratio test/reference)	98.5	88.23 – 109.98	38.85	99.8
AUC(0-tlast) (ratio test/reference)	88.68	81.55 – 96.42	29.08	>99.99

Figure 1. Mean plasma concentration-time curves after a single dose of a Test drug (Rabby-D 10 mg enteric coated tablet (gastro-resistant tablet), Neutecİlaç San. Tic. A.Ş., Turkey) containing 10 mg dexrabeprazole sodium and Reference drug (Pariet® 20 mg gastroresistant tablet, Eisai Limited European Knowledge Centre, United Kingdom) containing 20 mg rabeprazole sodium in healthy male subjects (N=36, 72 observations) under fasting conditions.

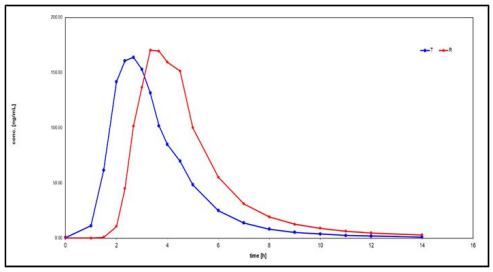
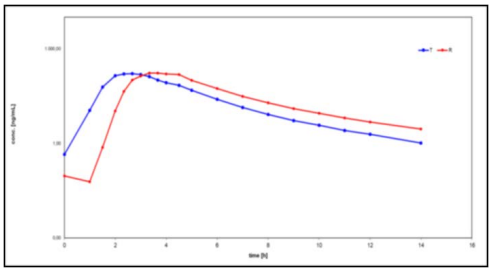
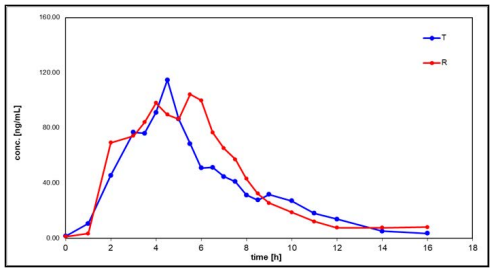
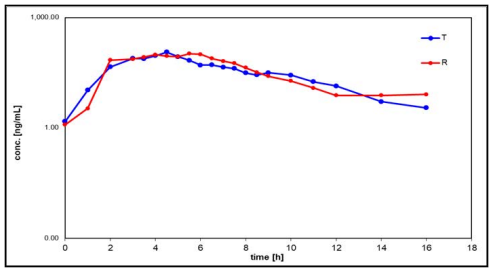
(A) Aritmetic  (B) Semi-logaritmic 

Figure 2. Mean plasma concentration-time curves after a single dose of a Test drug (Rabby-D 10 mg enteric coated tablet (gastro-resistant tablet), Neutecİlaç San. Tic. A.Ş., Turkey) containing 10 mg dexrabeprazole sodium and Reference drug (Pariet® 20 mg gastroresistant tablet, Eisai Limited European Knowledge Centre, United Kingdom) containing 20 mg rabeprazole sodium in healthy male subjects (N=34, 68 observations) under fed conditions.

(A) Aritmetic  (B) Semi-logaritmic 

ability for C_{max} is 22.68%; therefore a wider acceptance range for C_{max} wouldn't be considered; however 90% confidence limits for C_{max} was found within 80.00% to 125.00%. The conventional acceptance range (80.00% to 125.00%) for both $AUC_{(0\text{-tlast})}$ and C_{max} are acceptable.

The 90% confidence interval calculated for the primary endpoint, intra-individual ratios (T/R) for $AUC_{(0\text{-t})}$ of dexrabeprazole was 84.83% to 90.41%. The point estimator was 87.57%. The 90% confidence interval calculated for the primary endpoint, intra-individual ratios (T/R) for C_{max} of dexrabeprazole was 95.23% to 107.79% with a point estimator of 101.31%. Thus, the confidence intervals for $AUC_{(0\text{-tlast})}$ and C_{max} ratios were within the standard acceptance range of 80.00 – 125.00% under fasting conditions.

For fed conditions; bioequivalence evaluation was based on the geometric LSM ratios T/R of the primary endpoint parameters, $AUC_{(0\text{-tlast})}$ and C_{max} of dexrabeprazole. The 90% confidence interval calculated for the primary endpoint, intra-individual ratios (T/R) for $AUC_{(0\text{-tlast})}$ of dexrabeprazole was 81.55% to 96.42 and the point estimator was 88.68%. The 90% confidence interval calculated for the primary endpoint, intra-individual ratios (T/R) for C_{max} of dexrabeprazole was 88.23% to 109.98% with a point estimator of 98.50%. The intra-individual variability for C_{max} under fed conditions was 38.85%; therefore a wider acceptance range for C_{max} would be acceptable. However, as the 90% confidence intervals for both primary parameters were found within the standard bioequivalence acceptance range of 80.00% to 125.00%, a wider acceptance range was not required to be applied. Thus the confidence intervals for $AUC_{(0\text{-tlast})}$ and C_{max} ratios were within the standard acceptance range of 80.00 – 125.00% under fed conditions.

t_{max}swere analyzed using Wilcoxon test. There was a significant difference between two formulations with a significance level of 5% ($p<0.0001$) under fasting conditions and there was no significant difference between two formulations with a significance level of 5% ($p>0.05$) under fed conditions.

Conclusions

According to the Study Protocols, the $AUC_{(0\text{-t})}$ and C_{max} parameters for dexrabeprazole were used to assess bioequivalence. The results confirm that the 90% confidence intervals for Test to Reference ratios of the geometric least squares means for $AUC_{(0\text{-t})}$ and C_{max} were within the bioequivalence acceptance range of 80.00 to 125.00% under fasting and fed conditions.

The test product containing dexrabeprazole (Rabby-D 10 mg enteric coated tablet, NeutecİlaçSan. Tic. A.Ş.-Turkey) and reference product containing rabeprazole (Pariet® 20 mg gastro-resistant tablet, Eisai Limited European Knowledge Centre-UK)are bioequivalent in terms of rate and extent of absorption for dexrabeprazoleunder fasting and fed conditions. The two bioequivalence studies demonstrated equivalent system exposure of dexrabeprazole following oral dose given as dexrabeprazole alone (as test product Rabby-D 10 mg enteric coated tablet, NeutecİlaçSan. Tic. A.Ş.-Turkey) and as racemate rabeprazole (referenceproduct, Pariet® 20 mg gastro-resistant tablet, Eisai Limited European Knowledge Centre-UK). The test and reference formulations demonstrated similar tolerability. No adverse event and serious adverse event was registered during the studies. Both study drugs were well-tolerated and considered to be safe.

Acknowledgements

This study was funded by Genera Pharma AG., Wollerau, Switzerland. Clinical part of this study was conducted at Farmagen Good Clinical Practice Center, Gaziantep, Turkey, the bioanalyticanalysis were carried out by Novagenix Bioanalytical Drugs R&D, Ankara, Turkey and the biostatistical analysis were performed at TNC İlaçAraştırmaGeliştirmeDanışmanlık San. ve Tic. Ltd. Şti., İstanbul, Turkey.

Data Availability

The data that support the findings of this study are available from the corresponding author upon reasonable request. Some data may not be made available because of privacy or ethical restrictions.

References

[1]. Bodhankar SL, Jain BB, Ahire BP, Daude RB, Shitole PP.The effect of rabeprazole and its isomers on aspirin and histamineinduced ulcers in rats. Indian J Pharmacol 2006;38:357-358.
[2]. Pai V, Pai N. Randomized, double-blind, comparative study of dexrabeprazole 10 mg versus rabeprazole 20 mg in the treatment of gastroesophageal reflux disease. World J Gastroenterol. 2007 Aug 14;13(30):4100-2. PubMed PMID: 17696229.
[3]. Miura M, Satoh S, Tada H, Habuchi T, Suzuki T. Stereoselective metabolism of rabeprazole-thioether to rabeprazole by human liver microsomes. Eur J ClinPharmacol. 2006 Feb;62(2):113-7. PubMed PMID: 16389533.
[4]. Miura M, Kagaya H, Tada H, Uno T, Yasui-Furukori N, Tateishi T, et al. Enantioselective disposition of rabeprazole in relation to CYP2C19 genotypes. Br J ClinPharmacol. 2006 Mar;61(3):315-20. PubMed PMID: 16487225.
[5]. SPC, Electronic Medicines Compendium, PARIET 10 mg & 20 mg gastro-resistant tablet, Eisai Ltd, Last revised on 11/10/2017.
[6]. Guideline on The Investigation of Bioequivalence. CPMP/EWP/QWP/1401/98 Rev.1/Corr., London, EMA; 2010.
[7]. Guidance for Industry. Bioavailability and bioequivalence studies for orally administered drug products- General Considerations.FDA, CDER; 2014.
[8]. The Guidance for GCP, published by the Ministry of Health of Turkey. Circular, 13.11.2015.
[9]. Sun LN, Shen YW, Ying YW, Li D, Li TF, Zhang XH, et al. Stereoselective pharmacokinetics of (R)-(+)- and (S)-(-)-rabeprazole in human using chiral LC-MS/MS after administration of rabeprazole sodium enteric-coated tablet. Chirality. 2018 Dec;30(12):1277-1286. PubMed PMID: 30321480.
[10]. Guidance for Industry. Bioanalytical Method Validation.FDA, CDER; 2018.
[11]. Guideline on Bioanalytical Method Validation, EMEA/CHMP/EWP/192217/2009 Rev.1 Corr.2, London; 2011.