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DOSE RATIONALE BIOEQUIVALENCE ASSESSMENT UNDER FASTING CONDITIONS OF ELTROMBOPAG FOR BIOWAIVER MANUFACTURED IN TÜRKİYE

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ABSTRACT

Objective: *This study aimed to evaluate and compare the pharmacokinetics and tolerability of Eltrombopag 25 mg and 75 mg film-coated tablets (test product, Nobel İlaç) in healthy male volunteers with Revolade 25 mg and 75 mg film-coated tablets (reference product, Novartis Pharma GmbH, Germany) through two distinct studies, and the biowaiver of 50 mg dose from bioequivalence study by bracketing approach. Additionally, a bioequivalence waiver for Eltrombopag 50 mg was evaluated using a bracketing approach based on two bioequivalence studies (Study 1 and Study 2) from Eltrombopag (Test Product, Nobel İlaç) 25 mg and 75 mg Film-Coated Tablets.*

Material and Method: *Fasting studies for both doses were conducted using an open-label, randomized, single-dose, two-period, crossover design. A total of 46 and 32 volunteers were enrolled for the Eltrombopag 25 mg and 75 mg dose rationale studies, respectively. Participants were randomly assigned to two sequences (TR/RT) using a computer-generated randomization table.*

Results: *The geometric mean ratios of the Eltrombopag test and reference products fell within the 90% confidence interval as outlined in the Clinical Study Protocol, confirming bioequivalence. It was concluded that the test products, Eltrombopag 25 mg and 75 mg Film Coated Tablets (Nobel İlaç) test products are bioequivalent to originator formulation under fasting conditions.*

Conclusion: *Two distinct bioequivalence studies (Study 1 and Study 2) conducted for Eltrombopag 25 mg and 75 mg Film Coated Tablet doses were positively finalised and found to be bioequivalent to the reference product under fasting conditions. The biowaiver for the 50 mg dose was justified by dose proportional and bracketting approaches in accordance with Turkish Medicines and Medical Devices Agency (TMMDA) and European Medical Agency (EMA) guidelines. These findings support the safe and effective use of Eltrombopag within the therapeutic dose range.*

Keywords: *Eltrombopag, bracketing approach, bioequivalence, biowaiver, idiopathic thrombocytopenic purpura.*

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TÜRKİYE'DE ÜRETİLEN ELTROMBOPAG İÇİN AÇLIK KOŞULLARINDA DOZ ORANTISAL BİYOEŞDEĞERLİK DEĞERLENDİRMESİ İLE BİYOMUAFİYET YAKLAŞIMI

ÖZET

Amaç: Bu çalışmanın amacı, Eltrombopag 25 mg ve 75 mg Film Kaplı Tabletlerin (test ürün, Nobel İlaç) sağlıklı erkek gönüllülerde farmakokinetiğini ve tolerabilitesini Revolade 25 mg ve 75 mg Film Kaplı Tabletlerle (referans ürün, Novartis Pharma GmbH-Almanya) karşılaştırmalı olarak iki ayrı çalışma olarak incelemektir. Bu çalışma kapsamında Eltrombopag 50 mg Film Kaplı Tablet için biyoeşdeğerlik muafiyeti, Eltrombopag (Test Ürünü, Nobel İlaç) 25 mg ve 75 mg Film Kaplı Tabletlerden elde edilen iki biyoeşdeğerlik çalışmasına (Çalışma 1 ve Çalışma 2) dayanan bir bloklama yaklaşımı kullanılarak değerlendirilmiştir.

Materyal ve Metot: Her iki doz için açlık çalışmaları açık etiketli, randomize, oral tek doz, iki periyotlu, çapraz dizayn ile yürütülmüştür. Eltrombopag 25 mg ve 75 mg doz randomize çalışması için sırasıyla 46 ve 32 gönüllü çalışmaya katılmış olup, her iki doz için de gönüllüler bilgisayar kaynaklı randomizasyon tablosuna göre iki sıraya (TR/RT) randomize olarak kaydedilmiştir.

Bulgular: Eltrombopag'ın test/referans ürünlerinin geometrik ortalama oranları klinik çalışma protokolündeki %90 güven aralığında olup; yapılan biyoeşdeğerlik çalışmalarına göre Eltrombopag 25 mg ve 75 mg film kaplı tabletlerin (test ürün, Nobel İlaç) açlık koşullarında Revolade 25 mg ve 75 mg Film Kaplı Tabletlerle (referans ürün, Novartis Pharma GmbH–Almanya) ile biyoeşdeğer olduğu sonucuna varılmıştır.

Sonuçlar: Eltrombopag 25 mg ve 75 mg Film Kaplı Tablet dozları için yürütülen 2 ayrı biyoeşdeğerlik çalışması (Çalışma 1 ve Çalışma 2) olumlu sonuçlanmış olup, açlık koşullarında referans ürün ile biyoeşdeğer olarak bulunmuştur. 50 mg doz için yapılan muafiyet, TMMDA (Türkiye İlaç ve Tıbbi Cihaz Kurumu) ile EMA (Avrupa Tıp Ajansı) kılavuzlarına bağlı kalınarak doz orantısal ve bloklama yaklaşımlarıyla gerekçelendirilmiştir. Bu bulgular, Eltrombopag'ın terapötik doz aralığında güvenli ve etkin bir şekilde kullanımını desteklemektedir.

Anahtar kelimeler: Eltrombopag, bloklama yaklaşımı, biyoeşdeğerlik, biyomuafiyet, idiopatik trombositopenik purpura.

INTRODUCTION

Idiopathic thrombocytopenic purpura (ITP) is a hematologic disorder characterized by thrombocytopenia, moderate to severe bleeding, and an unidentified underlying cause.[1-3] ITP is associated with a decreased platelet count, the presence of normal megakaryocytes in the bone marrow, and enhanced platelet oxidation. Further, the concept of idiopathic denotes the exclusion of illnesses known to be linked to secondary thrombocytopenia. Given the severe and unpredictable nature of ITP, antithrombotic therapy plays a crucial role in preventing life-threatening complications such as venous thrombosis and pulmonary embolism. Eltrombopag, a newly developed thrombopoietin receptor agonist, appears to be effective in patients with idiopathic thrombocytopenic purpura.[4-7] There are many strategies to overcome distinctive comorbidities in cases. For this purpose, the primary strategy is treating patients with glucocorticoids and intravenous immunoglobulins to raise platelet counts in ITP by lessening the severity of platelet breakdown. However, current therapies that promote thrombopoiesis have resulted from the realization that platelet generation is insufficient in ITP. Among the drugs evaluated in the ITP scope, Eltrombopag is one of the active substances with compatible indications.[8]

The mechanism of action of the small molecule Eltrombopag, a non-peptide thrombopoietin receptor agonist, is to bind to the transmembrane domain of the thrombopoietin receptor on the surface of precursor cells. The main function of Eltrombopag is specific binding to the transmembrane domain of the thrombopoietin receptor on the surface of platelets, megakaryocytes, and megakaryocyte precursor cells. Megakaryocytes are large hematopoietic cells that makeup circulating platelets.[9-11] Eltrombopag, which is now accepted as an agent with potent thrombotic activity in first-line therapy, offers a new therapeutic modality by stimulating megakaryocyte proliferation and differentiation similar to the natural thrombopoietin system through the Kinase/Signal Transducer and Activator of Transcription (JAK/STAT) pathway.[12-14]

Both potential usage and easy access to molecules developed in pharmaceutical trends are subject to certain procedures and guidelines. In a light of European Medical Agency (EMA) guidelines, biowaivers can be

Table 1. Clinical routine laboratory practices.
Haematology
Serology
Serum Chemistry
Urinalysis
Hypotension (Systolic blood pressure (BP)≤100 mmHg or diastolic BP≤65 mmHg)
Hypertension (Systolic blood pressure (BP)≥150 mmHg or diastolic BP≥100 mmHg)

Table 2. The following formulations of test and reference product.

Drug Products	Manufacturer	Marketing Authorisation Holder
Test Drug-I: Eltrombopag 25 mg Film Coated Tablet	Nobel İlaç Sanayi ve Ticaret A.Ş.- Türkiye	Nobel İlaç Sanayi ve Ticaret A.Ş.- Türkiye
Reference Drug-I: Revolade 25 mg Filmtabletten	Novartis Pharma GmbH - Germany	Novartis Pharma GmbH - Germany
Test Drug-II: Eltrombopag 75 mg Film Coated Tablet	Nobel İlaç Sanayi ve Ticaret A.Ş. - Türkiye	Nobel İlaç Sanayi ve Ticaret A.Ş. - Türkiye
Reference Drug-II: Revolade 25 mg Filmtabletten	Novartis Pharma GmbH - Germany	Novartis Pharma GmbH - Germany

Table 3. Demographic data of volunteers for Study 1 (25 mg strength).

n=46	Age	Weight (kg)	Height (cm)	BMI (Kg/m²)
Mean	27.70	73.09	172.59	24.53
SD	9.31	9.70	6.18	2.94
Minimum	19	53	158	18.69
Maximum	50	91	187	29.75

SD: Standard deviation, **BMI:** body mass index

Table 4. Demographic data of volunteers for Study 2 (75 mg strength).

n=32	Age	Weight (kg)	Height (cm)	BMI (Kg/m²)
Mean	23.38	72.59	175.13	23.64
SD	3.74	10.33	5.88	2.96
Minimum	19	55	163	19
Maximum	36	91	189	29

SD: Standard deviation, **BMI:** body mass index

obtained with dose proportionality or multiple dose combinations. In this context, obtaining a biowaiver allows generic Research and Development (R&D) to increase access to the drug and avoid excessive costs. Obtaining biowaivers with such reliable methods prevents minimizing the numbers of volunteers and reduces clinical overcrowding. There are also economic benefits for sponsors.[15]

Herein, multiple-dose approaches within the scope of obtaining the antithrombotic effect is advance due to the variability of the responses in volunteer groups. According to EMA and Turkish Medicines and Medical Devices Agency (TMMDA) guidelines, to facilitate exemption for multiple dose studies biowaiver approaches are implemented. In this context, the pharmacokinetics and tolerability of Eltrombopag 25 mg and 75 mg Film-Coated Tablets (Nobel İlaç) in healthy male volunteers, to exempt 50 mg for Eltrombopag bioequivalence study. For this regard, separate doses with the innovator's conventional tablets, Revolade 25 mg and 75 mg Film Coated Tablets (Novartis Pharma GmbH) were examined.

MATERIAL AND METHOD

Volunteers

In this study, 25 mg (Study 1) and 75 mg (Study 2) Eltrombopag doses were evaluated as two different dosage studies for bracketing approaches. For this purpose, in the first study, 46 healthy Caucasian male volunteers aged between 19-50 years were assigned for Eltrombopag 25 mg Film Coated Tablet thus, 42 volunteers completed the study. On the other hand, in Eltrombopag 75 mg Film Coated Tablet study, 32 healthy male were included and completed the study for clinical trials. For study 1, the mean age was 23.38 years (±3.74 years), the mean weight was 72.59 kg (±10.33 kg), and the mean height was 175.13 cm (±5.88 cm). For study 2, the mean age was 28.58 years (±9.68 years), the mean weight was 73.38 kg (±9.52 kg), and the mean height was 172.68 cm (±5.92 cm).

Furthermore, In vitro dissolution testing was conducted to evaluate the rate and extent of drug release from the finished dosage form, as well as factors influencing its in vivo performance. According to the $f2$ biowaiver criteria, comparable in vitro dissolution data must be indicated during the in vivo bioequivalence testing. Dissolution profile frequently be examined at various pH values such as pH 1.2, 4.5, and 6.8.

Pharmacokinetic variables were compared, and the results were evaluated according to the TMMDA and EMA's guidelines. Bioequivalence studies were conducted for 25 mg and 75 mg strengths.

In addition, clinical routine laboratory practices were conducted with clinical criteria in both studies (Table 1). The main exclusion criteria were clinically significant findings (hematology, serology, serum chemistry, and urinalysis); hypotension (systolic blood pressure (BP) ≤100 mm Hg or diastolic BP ≤65 mm Hg) or hypertension (systolic BP≥150 mm Hg or diastolic BP≥100 mm Hg). Prescription drugs with the potential to interact with Eltrombopag within 2 weeks before the study or non-prescription drugs within 1 week before the study; and the use of drugs that induce or inhibit drug metabolizing enzymes with the potential to interact with the study drug within 1 month before the study.

The preparation of this protocol was based on the Helsinki Declaration. Likewise, the study protocol obeyed with all applicable Turkish laws and regulations as well as all relevant Good Clinical Practices (GCP) and Good Laboratory Practices (GLP) standards that may have been set internationally. GCP and GLP following the Declaration of Helsinki were applied in the conduct of this study and the storage of important documents.

The application of study drugs was performed by authorized persons involved throughout the study to ensure the accuracy of drug administration. The procedure for administration of the study medication was approved by the Principal Investigator, as reported in the Case Report Form (CRF), and the presence of the drug in plasma was reconciled, as well (Erciyes University Bioavailability and Bioequivalence Research Ethical Committee, Türkiye).

Drug Products

Detailed information on the test and reference products used in the bioequivalence study is presented in Table 2.

Design for Fasting Studies at 25 mg and 75 mg of Eltrombopag

Within the concept of the two distinctive studies, subjects were assigned according to a cross-over study design (Drugs were administered via cross-over between Period I and Period II). For study 1 and study 2, 46 and 32 healthy male volunteers received 25 mg and 75 mg single oral dose tablets (either reference or test drug) following at least 10 hours overnight fasting in each period in an order determined by randomization.

Under fasting conditions, both reference and test drugs were administered within 240 mL of water to volunteers. The volunteers in the study were served with an evening meal, and a standardized lunch (total calorie ~ 1200 kcal) was given 4 hours after dosing in each period. It was decided that the caloric composition of the same meal was necessarily standardized dinner given 10 hours after dosing in each period (with a total of ~1200 kcal) after dosing in each period.

Blood Sampling

Drugs were administered via cross-over and blinded procedures

For both studies, a 6 mL venous blood sampling procedure was conducted at pre-dose and at 0.50, 1.00, 1.50, 2.00, 2.50, 3.00, 3.50, 4.00, 4.50, 5.00, 5.50, 6.00, 6.50, 7.00, 8.00, 10.00, 12.00, 24.00, 48.00, 72.00 hours post-dose. Blood samples for the test drug and reference drugs were collected into polypropylene tubes via an indwelling catheter placed in the forearm as 300 mL after dosing. The anticoagulant K_2EDTA was used in polypropylene tubes. Blood samples for pharmacokinetic analysis were transferred to polypropylene tubes after being collected and kept at 2-8°C for not more than 30 minutes.

Blood samples were centrifuged at 3,000 rpm, 4-6°C for 10 minutes and plasma was separated from each sample. For each tube, 1.5 mL of the sample was transferred to polypropylene tubes.

The demographic data of volunteers for the two studies were presented in Table 3 and Table 4, respectively.

Bioanalysis

The quantitative determination of Eltrombopag was performed from human plasma by LC-MS/MS. During this determination, plasma levels of Eltrombopag were validated for a calibration curve ranging from 50 ng/mL to 20,000 ng/mL (Novagenix Bioanalytical Drug R&D Centre, Ankara, Türkiye) and data were presented by tandem mass spectrometry coupled to a validated liquid chromatography (LC-MS/MS) method. The best-fit range of the calibration standards in the presence of interpolation was determined and proposed with 8 calibration levels that cover the expected concentration range of the study. In addition, the parameters given in the table were successfully verified. Furthermore, analyte stability such as matrix effect, was evaluated and the data presented in Table 5.

Table 5. Analytical method validation parameters.	
Selectivity	√
Linearity	√
Precision	√
Accuracy	√
Dilution integrity	√
Stability	√
Effect of hemolyzed hyperlipiddemic plasma	√
Drug-drug interaction	√
Matrix Effect	√
Carry-over	√
Recovery	√
Reinjection reproducibility	√
Batch Size	√

Table 6. Pharmacokinetic parameters for 25 mg of Eltrombopag.

Parameter (Units)	Test (T) Arithmetic Mean±SD	Reference (R) Arithmetic Mean±SD
C_{max} (ng/mL)	3299.065±801.501	3116.908±925.689
$AUC_{0\text{-last}}$ (ng.hr/mL)	30667.752±11265.590	29207.690±11941.294
$AUC_{0\text{-}\infty}$ (ng.hr/mL)	32522.105±11889.322	31169.612±12632.753
t_{max}	3.000(1.500-4.000)	2.500(1.500-5.000)
$T_{1/2}$	13.857±5.584	13.945±5.315

SD: Standard deviation, **C_{max}:** maximum concentration, **$AUC_{0\text{-last}}$:** area under the plasma concentration-time curve from zero to last measurable concentration, **$AUC_{0\text{-}\infty}$:** area under the curve from zero to time infinity, **T_{max}:** time to peak drug concentration. **$T_{1/2}$:** half-life

Table 7. GeoLSM, ratios and 90% Confidence Interval (CIs) for Eltrombopag.

Parameter	TEST GeoLSM	REF GeoLSM	Ratio	90% CI	CV(intra) %	Power %
C_{max}	3195.8834	2940.6971	108.6778	99.9015-118.2249	22.9130	86.5623
$AUC_{0\text{-last}}$	28380.7140	26606.9880	106.6664	97.4063-116.8068	24.7647	89.3476

GeoLSM: Geometric least square means, **C_{max}:** Maximum concentration, **$AUC_{0\text{-last}}$:** area under the plasma concentration-time curve from zero to last measurable concentration, **CV:** coefficient of variation.

Table 8. Pharmacokinetic parameters for 75 mg of Eltrombopag.

Parameter (Units)	Test (T)	Reference (R)
C_{max} (ng/mL)	8228.013±1959.535	7269.465±1984.972
$AUC_{0\text{-last}}$ (ng.hr/mL)	70082.691±21326.956	64478.769±20718.795
$AUC_{0\text{-}\infty}$ (ng.hr/mL)	73680.980±23367.273	67767.867±22015.405
t_{max}	3.031±0.792	3.156±0.712
$T_{1/2}$	16.766±5.195	16.992±5.065
λ_z (1/h)	0.047±0.020	0.046±0.018
MRT(hr)	17.360±4.305	17.324±3.958

Arithmetic Mean ± SD (n=32)
SD: Standard deviation, **MRT:** mean residence time, **C_{max}:** maximum concentration, **$AUC_{0\text{-last}}$:** area under the plasma concentration-time curve from zero to last measurable concentration, **$AUC_{0\text{-last}}$:** area under the curve from zero to time infinity, **T_{max}:** time to peak drug concentration. **$t_{1/2}$:** half-life

The result of this validation showed that Eltrombopag can be analyzed in human plasma in an interval of 50-20,000 ng/mL according to the method described in Novagenix's Standard Operating Procedure document with reliability for pharmacokinetic studies. The validated LC-MS/MS method was implemented to determine Eltrombopag in plasma samples generated during the clinical phase of this study: 'Open-label, randomized, single oral dose, two-period, cross-over trial to assess the bioequivalence of Eltrombopag 25 mg Film Coated Tablet and Eltrombopag 75 mg Film Coated Tablet (Test Drugs, Nobel İlaç) in comparison with Revolade 25 mg Filmtabletten and Revolade 75 mg Filmtabletten (Reference Drug, Novartis Pharma GmbH-Germany) in healthy male volunteers under fasting conditions in two separate studies'.

All validation assays were presented with the Guidance for Bioanalytical Method Validation, EMA, International Council for Harmonisation (ICH) and the Food and Drug Administration (FDA) Guidance for Industry, Bioanalytical Method Validation. In the context of FDA and EMA guidelines, accuracy and precision were within the desired ranges in the presence of implemented criteria.

Tolerability Assessment

Volunteers participating in the studies are required to have an initial follow-up examination at least 14 days in advance. The initial examination consisted of a procedure including demographic data, a standardised clinical assessment, a brief history and clinical findings. All

The standard laboratory screening analyses for both studies are presented in Figure 1. In this context, a total of 24 mL of blood samples (12 mL of input and 12 mL of final) were collected from each volunteer under fasting conditions for laboratory examinations in standard laboratory screening. Figure 1 also presents the parameters determined from 30 mL of urine. In addition, the researcher was provided with a certified printout of the original laboratory values.

Pharmacokinetic and Statistical Analysis

In pharmacokinetic analysis, plasma concentration of Eltrombopag 25 mg and 75 mg were used to determine within the following parameter; C_{max}, $AUC_{0\text{-tlast}}$, $AUC_{0\text{-}\infty}$, t_{max}, $t_{1/2}$, MRT, λz.

The statistical analysis for 25 mg Eltrombopag was conducted via Phoenix WinNonlin (Version 8.3.5, Certara L.P.) on the other hand, for 75 mg Eltrombopag

was analyzed implementing Phoenix WinNonlin (Version 8.1, Certara L.P.). Furthermore, Analysis of Variance (ANOVA), two one-sided tests, and 90% confidence intervals for geometric mean Cmax and AUC0-tlast ratios were calculated using both 25 mg and 75 mg Eltrombopag plasma concentrations. Safety assessments were based on analyses of separate laboratory values and descriptive statistics in each of the two treatments for both dose studies.

For Eltrombopag 25 mg and 75 mg Film Coated Tablets, relative comparison in the bioavailability analysis was conducted at 90% Confidence Intervals (CIs) for geometric mean ratios (test versus reference drug) within the range of 0.80 to 1.25 for the primary pharmacokinetic parameters (Cmax and AUC0-tlast for the fasting studies) following Türkiye Medicines and Medical Devices Agency (TMMDA) 5and international guidelines (ICH, EMA and FDA Guidelines).[15-17]

Pharmacokinetic data of volunteers for both 25 mg and 75 mg fasting studies in Table 6, Table 7, Table 8, and Table 9, respectively.

RESULTS

Eltrombopag 25 mg (Study 1);

A cross-over design was used in the bioequivalence study. This procedure is intended to increase the efficiency of the test from the statistical point of view of the cross-over. Although the study was open-label, the allocation of the test and reference products was not known by CRO until the analytical part of the study completed.

58 volunteers were screened. 46 volunteers were randomly assigned into two sequences (TR/RT). At the end of the study, 42 volunteers finalized the clinical phase of the study within the planning process. There have been four drop-outs. Subjects 11 and 13 left the study with their own decisions at the first isolation day of Period I and the hospitalization day of Period I, respectively. Subjects 16 and 17 did not come to the second period isolation day of Period II. They have not been replaced following the Clinical Study Protocol. Test product was administered to 42 completed volunteers and Reference product was administered to 42 completed volunteers. Two drop-outs (Subject 11 and Subject 13) did not take any product and two drop-outs (Subject 16 and Subject 17) took only Reference drug in Period I. After the 10-day washout period; The Eltrombopag was administered by the subjects in Period I and Period II by vice versa (Figure 2).

Table 9. Geometric means of 75 mg Eltrombopag, ratios and 90% confidence intervals.

Parameter	Difference	DiffSE	TESTLSM	REFLSM	Ratio	90%CI	CV%	Power%
ln(C_{max})	0.1293	0.0521	8.9846	8.8553	1.1380	1.0417-1.2433	21.0773	54.5671
ln(AUC_{0-last})	0.0808	0.0555	11.1040	11.0232	1.0842	0.9866-1.1913	22.4893	80.4945

ln (C_{max}): Logarithmic maximum concentration, **ln (AUC_{0-last}):** Logarithmic area under the plasma concentration-time curve from zero to last measurable concentration, **CI:** confidence interval, **CV:** coefficient of variation,

Regarding 25 mg dose, Cmax for Test and Reference products were demonstrated as 3,299.065±801.501 ng/mL and 3116.908±925.689 ng/mL, respectively. Besides, the mean ± SD of AUC0-tlast for Test and Reference products were 30,667.752±11,265.590 hr.ng/mL and 29,207.690±11,941.294 hr.ng/mL, whereas median (min-max) of tmax for Test and Reference products were 3.000 (1.500-4.000) hr and 2.500 (1.500-5.000) hr, respectively. Overall,

Figure 1. Screening parameters.
eGFR: Estimated Glomerular Filtration Rate, **SGOT (AST):** serum glutamate-oxaloacetate transaminase (aspartate aminotransferase), **SGPT (ALT):** serum glutamate-pyruvate transaminase (alanine aminotransferase, **GGT:** gamma-glutamyl transferase, **ECG:** electrocardiogram.

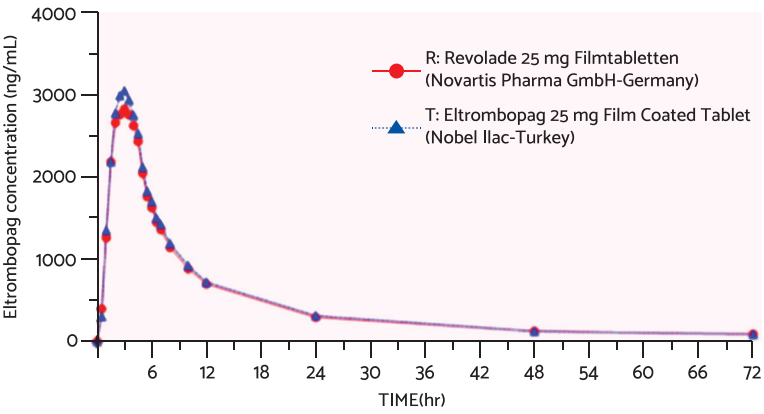

Figure 2. Average Eltrombopag 25 mg plasma concentration-time curve for fasting study.

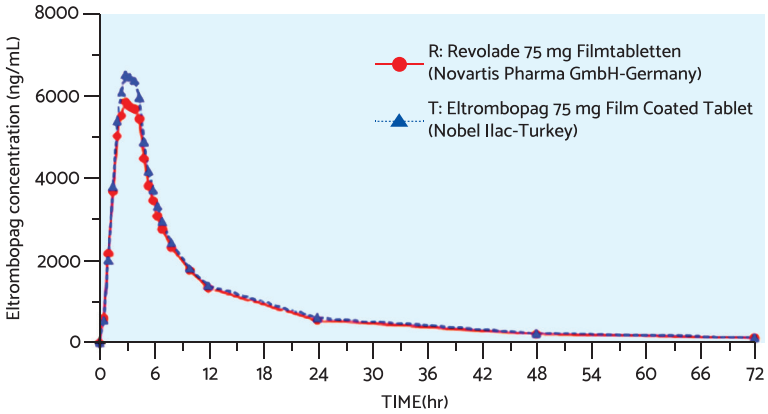

Figure 3. Average Eltrombopag 25 mg plasma concentration-time curve for fasting study.

Tmax was analyzed using a nonparametric approach, the Mann–Whitney U test. There was no significant difference between the two formulations regarding t_{max} (p>0.05). The mean ± SD of $t_{1/2}$ for Test and Reference product were 13.857±5.584 hr and 13.945±5.315 hr, respectively. $t_{1/2}$ for test and reference products ranged from 4.552 hr to 31.394 hr and 6.234 hr to 27.818 hr, respectively.

The intra-subject variabilities of C_{max} and $AUC_{0\text{-}tlast}$ have been found as 22.9130% and 24.7647%, respectively and the power related C_{max} and $AUC_{0\text{-}tlast}$ has been found as 86.5623% and 89.3476%, respectively (Table 7).

Eltrombopag 75 mg (Study 2)

In this procedure, 63 volunteers were screened. Thus, 32 volunteers were randomized and included in Study 2. During the study, there were no drop-out. As a result, 32 volunteers completed the clinical phase of the Study 2.

At the end of the clinical study period, oral examinations, medication accountability (used and unused), and bioanalysis were performed to assess treatment compliance. All research medications have been given out under the direction of the investigator or one of his agents.

The descriptive pharmacokinetic data C_{max} (mean±SD) for the Test and Reference products were found as 8,228.013±1,959.535 ng/mL 7,269.465±1,984.972 ng/mL, respectively. The mean $AUC_{0\text{-}tlast}$ was found as 70,082.691±21,326.956 hr.ng/mL for the Test product and 64,578.769±20,718.795 hr.ng/mL for the Reference product (Figure 3).

The mean±SD of t_{max} for Test and Reference products were 3.031±0.792 hr and 3.156±0.712 hr, respectively. T_{max} ranged from 1.50 hr to 4.50 hr for the test product and from 2.00 hr to 4.50 hr for the reference product.

Within the nonparametric test (Mann-Whitney U test), T_{max} was examined using a at 5% significance level. There were no crucial distinctions ($p>\alpha$) between the two formulations concerning t_{max}. $t_{1/2}$ for Test and Reference products were found as 16.766±5.195 hr and 16.992±5.065 hr, respectively. For the elimination rate of both contents, $t_{1/2}$, for test and reference products ranged from 6.490 hr to 28.929 hr and 6.548 hr to 25.198 hr, respectively. The mean± SD of λz for Test and Reference products were 0.047±0.020 hr^{-1} and 0.046±0.018 hr^{-1}, respectively. λz for test and reference products ranged from 0.024 hr^{-1} to 0.107 hr^{-1} and 0.028 hr^{-1} to 0.106 hr^{-1}, respectively (Table 8).

Tolerability

In the 25 mg Eltrombopag bioequivalence study: Eleven mild, possibly drug-related, and two mild, unlikely drug-related adverse events were reported during the clinical study. Five of the adverse events were fully recovered and one of them was recovered. Seven of the adverse events' results were not known.

In 75 mg Eltrombopag bioequivalence study: There have been seven mild adverse events were observed during the study. According to the results, the general safety of the products was in acceptable range, and Test and Reference products were safe in each study. The analysis of the safety and overall tolerability results showed that: Two Eltrombopag products were safe.

DISCUSSION

The perspective of this study is to examine the bioequivalence of the pharmacokinetic formulation developed by NOBEL ILAC in relation to the originator reference product, as part of implementing a biowaiver for the 50 mg dose of Eltrombopag. For this purpose, 42 and 32 healthy male volunteers completed the trials (Study 1 and Study 2, respectively) in accordance with bioequivalence studies following the TMMDA and EMA guidelines.

The results obtained at the end of the study showed that the Eltrombopag 25 mg and 75 mg Film Coated Tablet test products, are bioequivalent to the reference product in healthy Caucasian male volunteers based on the regular criteria for bioequivalence.

The developed formulation of Eltrombopag in 25 mg and 75 mg doses ensures efficacy in dosage proportional development potential, as well as in situations such as posology and safety. In posology studies aimed at reducing side effects that may occur in patient groups, Bosi *et al*. evaluated serum lactate dehydrogenase (LDH) levels after Eltrombopag treatment. It was reported

that LDH levels were 1.19 times the upper limit of normal (ULN) and total bilirubin levels were 1.19 mg/dL.[18] From a posology perspective, initial treatment with Eltrombopag includes a daily dose ranging from a minimum of 12.5 mg to 25 mg. The dose is increased to prevent bleeding until the platelet volume exceeds 50 cm^3. If the patient's platelet count is between 50 cm^2 and 200 cm^3, the dose should be maintained without change. If the platelet volume is over 200 cm^3, the treatment should be gradually reduced by 25 mg per day.[19]

Considering the findings obtained in the literature, it was observed that Eltrombopag tolerability was at a moderate level. However, when the patient data are compared in this context, adverse events arising from the initial phase of treatment were reported in 37% of patients during treatment with Eltrombopag. For the demonstrated study, there have been six possible drug-related adverse event and one unrelated adverse event was observed. According to the results, the tolerability of the administered drugs by the patients was found to be generally convenient.[20]

Furthermore, the minimum recommended dose for Eltrombopag is 50 mg. In our study, pharmacokinetic studies were performed for 25 mg and 75 mg, and bioequivalence data were presented by analyzing the tolerability with inter-dose comparison. In light of the data presented, it was planned to gain biowaiver from the bioequivalence study for 50 mg Eltrombopag prevent unnecessary clinical trials, and provide economic benefit by protecting patient health and safety. Within this therapeutic window, it is predicted that 50 mg Eltrombopag may be effective in obtaining a biowaiver and facilitating it on the market.[20]

CONCLUSION

Based on the findings from the bioequivalence studies, both Eltrombopag doses were determined to be bioequivalent to the reference drugs under fasting conditions, with an acceptable tolerability profile. According to the bracketing approach outlined in the TMMDA and EMA guidelines, a biowaiver was successfully justified. In this way, the implementation of biowaiver request as reliable methods tend to be prevent unnecessary volunteer utilization. Thus, it reduces the intensity in the clinic. Finally, with this study it purposed to utilize the economic portfolio for the sponsor and to prevent possible undesirable effects.

*The authors declare that there are no conflicts of interest.

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