Research Article

Clinical evaluation in adult human revealed the biosimilarity of recombinant Erythropoietin GBPD002 with Eprex®

Mamun Al Mahtab[1*], Sitesh Chandra Bachar[2*], Kakon Nag[4,5*], Mohammad Mohiuddin[4], Md. Abdur Rahim[1], Md. Helal Uddin[3], Samir Kumar[4], Md. Maksudur Rahman Khan[4], Md. Enamul Haq Sarker[4], Rony Roy[4], Sourav Chakraborty[4], Bipul Kumar Biswas[4], Md. Emrul Hasan Bappi[4], Ratan Roy[4], Uttam Barman[4] and Naznin Sultana[4,5*]

Abstract

The biosimilarity for erythropoietin (EPO) functionality of GBPD002 (test candidate) and Eprex® (comparator) has been evaluated by comparing the pharmacokinetic (PK) and pharmacodynamic (PD) properties following subcutaneous injection. This was a randomized, double-blinded, two-sequence, crossover clinical trial. Subjects were randomly assigned and received a dose (4,000 IU) of either the test or comparator EPO, and received the alternative formulations after 4-weeks of washout period. The PK parameters, *viz.*, maximum observed concentration (C_{max}) and area under the curve extrapolated to infinity (AUC_{0-inf}), were calculated with the serum EPO concentrations from blood samples and were found comparable for both formulations. The geometric mean ratios (at 90% CI) of the C_{max} and AUC_{inf} were 0.89 and 1.16, respectively, which were within the regulatory range of 0.80 – 1.25. The time-matched serum EPO concentrations and PD markers (reticulocyte, hematocrit, hemoglobin, and red blood cell) denoted a counter-clockwise hysteresis, suggesting a time delay between the observed concentration and the response. ANOVA-derived *p*-value (>0.05) for the effectors clearly revealed the similarity between effects on PD markers for the test and comparator drugs. Both formulations were found tolerated well, and anti-drug antibodies were not observed. Thus, the two formulations are projected to be used interchangeably in clinical settings.

Affiliation:
[1]Department of Hepatology, BSMMU, Dhaka, Bangladesh.
[2]Faculty of Pharmacy, University of Dhaka, Dhaka, Bangladesh.
[3]Clinical Research Organization Ltd. (CRO Ltd.), Dhaka, Bangladesh.
[4]Globe Biotech Limited, 3/Ka (New) Tejgaon I/A, Dhaka 1208, Bangladesh.
[5]R&D Management Solution Inc., Hamilton, Ontario L9C2V8, Canada.

*Corresponding author:
Mamun Al Mahtab, Department of Hepatology, BSMMU, Dhaka, Bangladesh, **Email:** shwapnil@agni.com. Sitesh Chandra Bachar, Faculty of Pharmacy, University of Dhaka, Dhaka, Bangladesh, **Email:** bacharsc63@gmail.com. Kakon Nag, Globe Biotech Limited, 3/Ka (New) Tejgaon I/A, Dhaka 1208, Bangladesh, **Email:** kakonpoly@yahoo.com. Naznin Sultana, Globe Biotech Limited, 3/Ka (New) Tejgaon I/A, Dhaka 1208, Bangladesh, **Email:** kakonpoly@gmail.com.

Keywords: Erythropoietin; Biosimilarity; Pharmacokinetics (PK); Pharmacodynamics (PD); Toxicity; Safety

Introduction

Erythropoietin (EPO) is a glycoprotein hormone that plays a key role in the formation of red blood cells (RBCs) [1]. EPO is primarily synthesized in the peritubular cells of the kidney and released into the systemic circulation in adult individuals [2]. Circulating EPO binds to the EPO receptor on bone marrow erythroid progenitors, triggering multiple signaling pathways that support differentiation into mature RBCs [3]. A reduction in EPO production is the primary cause of anemia in people with chronic renal failure [4]. Human recombinant epoetin (rHuEPO) or erythropoiesis-stimulating agents (ESA) have been demonstrated to stimulate erythropoiesis in anemic patients with chronic renal failure, including those who need and don't need dialysis [5-7]. Hereafter, EPO and rHuEPO have been used in this article interchangeably. ESAs are used to treat chemotherapy-induced anemia in cancer patients and to reduce the requirement for allogenic blood transfusions in patients with

mild anemia who are undergoing surgery [8-10]. Furthermore, EPO is recommended for patients who are at high risk for perioperative transfusions due to considerable blood loss. Alpha epoetins are the most commonly used type of rHuEPO among the other forms. Despite its tremendous importance in the clinical field, the price of EPO remains significantly high, and limiting its availability to the mass people – particularly, to the underdeveloped and developing countries. Eprex®, the pioneer product of alpha epoetins, is a regular medicine with proven efficacy and tolerability [11]. Eprex® was manufactured by Johnson & Johnson and it was the first EPO formulation to receive regulatory approval in Europe in 1988. In the early 1990s, physicians outside of the United States adopted the subcutaneous route of administration of EPO for hemodialysis patients due to the socio-economic benefit for the patients [11]. Human serum albumin (HSA), the stabilizer in Eprex® formulation, was changed to a synthetic compound, polysorbate 80, due to the concerns that albumin might transmit Creutzfeldt-Jakob disease [11]. Subsequently, HSA-free Eprex® has been available in market. GBPD002 is a biosimilar of Eprex®, which is developed by Globe Biotech Limited and synthesized in genetically engineered Chinese hamster ovary (CHO) cells. Upstream and downstream process development and validation were done for large-scale production [12]. Step- by-step analytical results confirmed the biosimilarity of GBPD002 with Eprex® regarding molecular characterization [13]. Single and repeat-dose toxicity were performed in Wister rats to analyze the toxicity of GBPD002 with Eprex® and were found safe for administration. PK/PD studies were performed in animal models and results were found similar for GBPD002 and Eprex® [13]. Here, the aim of this study is to analyze the bioequivalence of GBPD002 and Eprex®. The purpose of this study is to compare the pharmacokinetic (PK), pharmacodynamics (PD), and safety of rHuEPO, GBPD002, developed by Globe Biotech Limited, and the reference product Eprex® manufactured by Janssen Cilag Ltd., UK in healthy adult volunteers.

Material and methods

Study design

A randomized, double-blind, single-dose, and two-sequence crossover trial in healthy volunteers was designed. The protocol for the study has got ethical clearance from the institutional review board (IRB; The Ethics Committee of Farabi General Hospital Ltd.) and was approved by the Directorate General of Drug Administration (DGDA) of Bangladesh [14]. The Clinical Research Organization (CRO Ltd.) conducted this investigation at Farabi General Hospital, Dhanmondi R/A, Dhaka 1209, in compliance with the principles of the Declaration of Helsinki and the International Conference on Harmonization's Guideline for Good Clinical Practice. The informed consent form (ICF), along with the

protocol and methodology, was also approved by IRB and DGDA. Volunteers were given thorough information about the risks and benefits of the study and they signed the ICF to affirm their willingness to voluntarily participate in the study. The trial protocol has been submitted and registered with the clinicaltrial.gov of the National Library of Medicine of the USA [15]. Briefly, the study was open to healthy male participants aged 18 – 45 years old who weighed 55.0 – 90.0 kg and had a body mass index (BMI) of 18.0 – 27.0 kg/m2. Subjects were eliminated if they had at least one of the following clinical laboratory test results: hemoglobin level <12 g/dL or >17 g/dL, vitamin B12 level <200 pg/mL, ferritin level <21.8 ng/mL, transferrin level <190 mg/dL and any anomalous range for the reticulocyte (RET) count, erythrocytes, platelets or serum potassium levels. The study design is supported by a previously accomplished similar study [16]. The total number of subjects was 42, assuming a 20% dropout rate. HIV, HBsAg, and HCV (Hepatitis C Virus) positive individuals were excluded from the study. The study was conducted after the emergence of COVID-19, and therefore, proper precautions have been taken to protect the volunteers against exposure from COVID-19 during the trial. For example, during the primary selection, admission into the trial site, interaction during sampling and monitoring (including periods for waiting, washroom usage, food and drink intake, entry and exiting, housekeeping, etc.) were strictly controlled. The usage of approved masks and hand sanitizers was mandatory during the stay at the trial facility. All relevant people including investigators, medical doctors, nurses, analysts, support staffs, etc. who were engaged in the trial and with an opportunity to access the trial site were controlled; all of them followed the same rigorous access and mobility protocol. The volunteers were housed in isolated cabins where maximum of 2 persons were allocated in a single room in separate beds located at a minimum of 6 feet distance to reduce the frequency of interaction between subjects and interacting people. Regular COVID-19 tests were included for relevant sampling points, and the subjects with positive test results were immediately isolated and subjected to proper medical care. The recruited volunteers were randomly assigned to one of the two sequences (Sequence/Group A and Sequence/Group B) and received a single subcutaneous injection of 4,000 IU of either the comparator/reference drug (Eprex®) or the test drug (GBPD002) in the abdomen area from a single-use prefilled syriQ BioPure® syringe (SCHOTT Schweiz AG, Switzerland). The allocated sequences with a 28-day washout period were as follows: Group A, administered the comparator drug in period 1 followed by the test drug in period 2; Group B, given the test drug in period 1 followed by the comparator drug in period 2. Blood samples were taken for the PK evaluation at pre-dose and at 1, 3, 6, 8, 10, 14, 24,

48, 72, 96, 120, and 144 h post-dose. For the PD evaluation, the reticulocyte count (RET, %), hematocrit (HCT, %), haemoglobin (HB) (g/L), and red blood cell (RBC) count (10^6/mm³) were calculated at pre-dose and at 72, 144, 216, and 312 h post-dose. To maintain iron supply, instead of iron supplements in the form of medicine, the subjects were given a standard portion of iron-rich food prepared with green leafy vegetables, animal organs like the liver and red meat, beans, pumpkins, watermelon, dried dates and, dark chocolates were provided as snacks.

Bioanalytical method

A validated enzyme-linked immunosorbent assay (ELISA) technique was used to measure serum EPO quantities. *Quantikine® IVD® ELISA* , a human EPO immunoassay kit (R&D Systems Inc., USA) was used to determine the serum EPO concentrations. Exogenous EPO derived from rHuEPO and endogenous EPO were measured together in the same way. The procedure was validated following international guidelines. The calibration curve was constructed using seven distinct concentrations of calibration standard samples. Samples of low to high concentrations (2.5, 5, 20, 50, 100, and 200 mIU/mL) were prepared for quality control. Calibration curves of the test and comparator drugs showed linearity, ($r^2 > 0.99$ for both test and comparator) within the concentration range of 2.5 – 200 mIU/mL. The hematologic parameters for PD assessment (RET, HCT, HB, and RBC count) were analyzed in a diagnostic center (Lab Science Diagnostic, Bangladesh), which is accredited by the Directorate General of Health Services (DGHS) of Bangladesh.

PK and PD analyses

The PK parameters were measured by a non-compartmental method using Phoenix WinNonlin™ Version 8.3 software (Certara, USA). Raw data were used to determine the maximum observed serum EPO concentration (C_{max}) and the time of C_{max} (T_{max}). The last observed area under the curve (AUC_{last}) was determined by the linear trapezoidal method up to T_{max} and by the log trapezoidal method after T_{max}. The area under the curve was extrapolated to infinity (AUC_{inf}) with the following formula: $AUC_{inf} = AUC_{last} + C_{last/\lambda z}$, where C_{last} refers to the last observed serum EPO concentration and λ_z refers to the estimated terminal elimination rate constant. The terminal half-life ($t_{1/2}$) was calculated by dividing natural-log 2 by λ_z. The total clearance (CL/F) was determined with the following formula: CL/F = dose/AUC_{last}, where F means the bioavailability. The mean residence time (MRT_{last}) was calculated by dividing the area under the first moment curve by the AUC_{last} [14]. Goodcalculator™ and socscistatistics™ were used respectively to determine 90% CI and *p*-value. As PD indicators, the time courses of RET count, HB, HCT, and RBC count were studied and compared between the test and comparator drugs. The linear trapezoidal approach was used to determine the maximum effect change (E_{max}) and the area under the baseline-adjusted effect curve (AUEC) for the RET count, HB, HCT, and RBC count using baseline-adjusted values. We analyzed inter-and intra-volunteer data trends for all parameters, and outlier data were excluded from the final analysis to minimize errors in data prediction [17]. The time-matched PK/PD data (serum EPO concentration and each PD marker) were also plotted on a scatter plot to investigate the PK/PD time delay.

Safety and tolerability analysis

Safety and tolerability profiles of the drugs were evaluated in participants who had at least one dose of the study drug. The results of vital sign evaluations, electrocardiograms, and clinical laboratory testing were used to determine the safety and tolerability of the drug. Local reactivity for drug administration was assessed 1, 24, and 48 hours after injection. Anti-drug antibody (ADA) production was measured at the pre-dose of each period and at the post-study visit to determine the immunogenicity of the study drugs.

Statistical analysis

The key PK parameters C_{max} and AUC_{inf} were used in the PK comparison. A linear mixed-effect analysis of variance was used to calculate the log-transformed C_{max} and AUC_{inf}, with a fixed effect for the formulation, period, and sequence and a random effect for the subject nested for the sequence. For each PK parameter, the geometric mean ratio (GMR) of the test to the comparator was determined, along with its 90% CI. If the 90% CI for each PK parameter was within the range of 0.80 – 1.25, the test drug was determined to have PK equivalence with the comparator drug. The key PD parameters, E_{max} and AUEC of the RET count, were included in the PD comparison. The mean difference between the test and the comparator drug was calculated using the linear mixed-effect analysis of variance, along with its 90% CI and *p*-value. Statistical significance was defined as a *p*-value of less than 0.05; the *p*-value(s) for PD marker(s) for the EPO formulations greater than 0.05 were considered similar and non-significant.

Results

Demographics

Total of 83 persons were screened to include 42 volunteers (50.6% were eligible). The number of the study subject has been aligned with several other studies to achieve significant data collection and confident decision-making [17 – 21]. It has been shown that comparatively a lower dose provides more stable PK/PD results than the higher dose [22, 23]. Several studies have administered 4,000 IU dose for bioequivalence

studies as an effective and comparably low dose, [16, 24], and accordingly, we have used 4,000 IU/subject as the experimental dose. One volunteer from group A (started with comparator drug) and two volunteers from group B (started with test drug) dropped out from the study after the first phase (28 days) and before receiving the second dose due to being positive for COVID-19. The mean ± standard deviation (SD) [min–max] for age, height, weight, BMI, systolic and diastolic blood pressure of the volunteers were measured (Table-1). The demographic and other baseline variables were not found significantly different among the two groups.

PK results

After a single subcutaneous injection of both formulations, the amplitudes of serum EPO concentrations followed comparable time-dependent distributions (Supplementary Table-1 and 2). EPO demonstrated a delayed systemic absorption with a median T_{max} of $6 - 8$ h in both formulations and displayed multiphasic characteristics in the elimination phase (Figure-1). The GMR (test/comparator) for both the C_{max} and AUC_{inf} fell within the pre-specified range of 0.80–1.25 (0.89 and 1.16, respectively), suggesting that the two epoetin alfa formulations have similar PK profiles. The remaining PK characteristics were similar between the two formulations (Table-2).

PD results

RET, HB, HCT, and RBC were counted (Supplementary Table-3 – 6) after subcutaneous administration of the test or the comparator drug products. The mean RET counts progressively increased up to 72 hours following drug administration and declined until the last observation time (at 336 h). The test and comparator EPO had a similar rate of RET count change over the observed time span. The key PD parameters, viz., mean E_{max} and AUEC of the RET count, were comparable between the test and the comparator (p-value =0.604 and 0.976, respectively). Furthermore, count changes, E_{max}, and AUEC were similar for HB, HCT, and RBC between the two formulations (Figure-2 and Table-3). The similarities in PD parameters between the comparator and the test drug were clearly evident from the non-significant p-values, which are above 0.05 for each PD parameter.

Safety and tolerability

After receiving a single subcutaneous injection of 4,000 IU EPO, 27 subjects reported 88 adverse events (AE) (Table-4). The AEs were addressed immediately by the medical team as per protocol and recorded. Between the two treatments, the number of patients with AEs and the number of AEs were comparable. All the treatment-related AEs were mild in severity and did not require any medication. General weakness and headache were the most commonly reported treatment-related AEs, which are already recognized to be common mild side effects of rHuEPO formulations [25]. Clinical laboratory results, ECG readings, vital signs, and physical tests all showed no clinically significant changes for receiving two doses (test and comparator) of EPO. No local reaction was observed on the injection site among the volunteers. No ADA reactivity was found in any samples from both of the treatment groups.

Table 1: Demographic characteristics of the study groups.

Demographic characteristics	Sequence/Group A; (*n*=21)	Sequence/Group B; (*n*=21)
Mean age (years)	27.67±6.03	31.57±6.90
Mean height (inch)	64.4±0.21	64.1±0.17
Mean weight (kg)	59.94±6.50	64.88±6.00
Mean BMI (kg/m²)	22.4±2.21	24.5±3.35
SBP (mmHg)		
Baseline	115.5±5.41	122.78±7.45
Endpoint	115.6±6.96	123.13±10.24
DBP (mmHg)		
Baseline	75.0±4.26	79.44±1.49
Endpoint	73.20±6.04	81.13±5.09
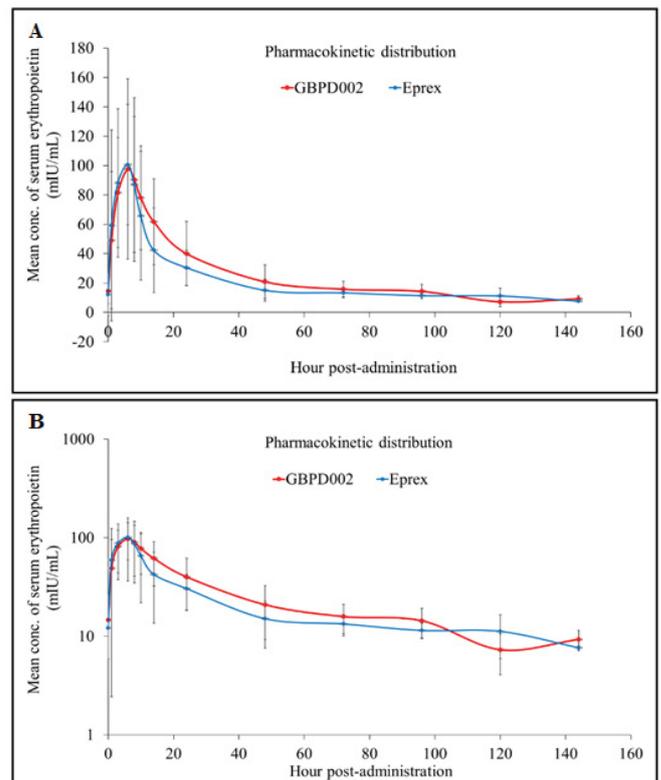
Figure 1: Two EPO formulations [comparator (Eprex®, blue line) and test (GBPD002, red line)] showed similar mean concentration of serum EPO profile with time after a single subcutaneous injection where (A) liner numerical scale, (B) log10 scale.

Citation: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Mohammad Mohiuddin, Md. Abdur Rahim, Md. Helal Uddin, Samir Kumar, Md. Maksudur Rahman Khan, Md. Enamul Haq Sarker, Rony Roy, Sourav Chakraborty, Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Uttam Barman and Naznin Sultana. Clinical Evaluation of Recombinant Erythropoietin Biosimilar GBPD002 Compared with Eprex® in Adult Human. Archives of Clinical and Biomedical Research. 7 (2023): 447-462.

Table 2: Pharmacokinetic parameters after a single subcutaneous administration of the test or the comparator drug products.

Parameters	Eprex®; (*n*=41)		GBPD002; (*n*=40)		GMR (90% CI)[a]
	Mean ± SD	CV	Mean ± SD	CV	
T_{max} (h)[b]	6.56±2.63	0.4	7.05±2.89	0.41	-
C_{max} (mIU/mL)	141.18±49.41	0.35	125.21±53.34	0.42	0.89
$AUC_{0-144 h}$ (h.mIU/mL)	2714.37±482.88	0.18	3299.37±833.63	0.25	1.22
AUC_{0-inf} (h.mIU/mL)	3381.29±424.3	0.13	3908.72±812.01	0.21	1.16
$t_{1/2}$ (h)	17.88±3.66	0.56	19.31±2.69	0.29	-
CL/F (mL/h)	619.98±22.81	0.04	656.31±61.48	0.37	-
MRT_{last} (h)	93.62±52.81	0.56	87.17±24.31	0.28	-
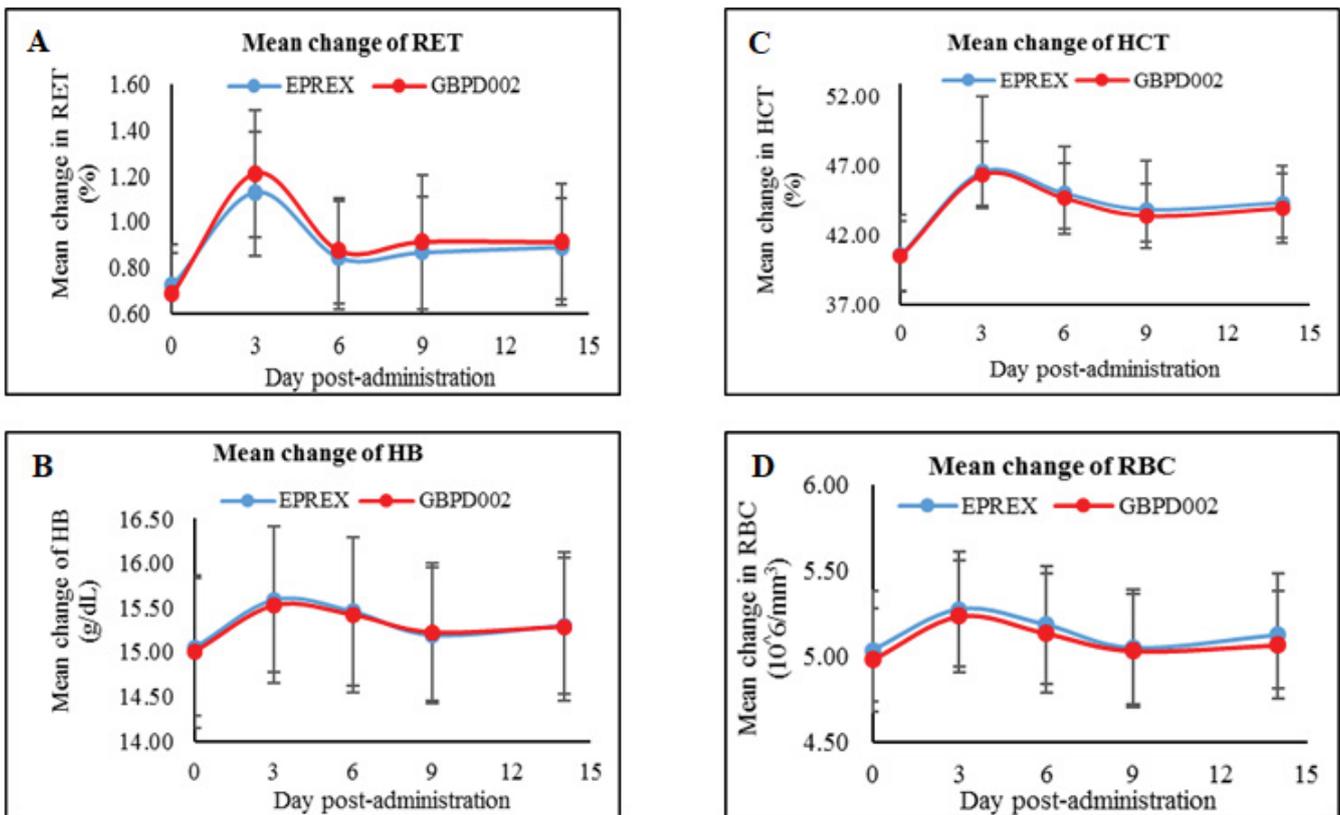
Figure 2: Two EPO formulations [comparator (EPREX®, blue line) and test (GBPD002, red line)] showed similar mean changes for hematologic parameters with time following a single subcutaneous administration where (A) RET, (B) HB, (C) HCT and (D) RBC.

Citation: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Mohammad Mohiuddin, Md. Abdur Rahim, Md. Helal Uddin, Samir Kumar, Md. Maksudur Rahman Khan, Md. Enamul Haq Sarker, Rony Roy, Sourav Chakraborty, Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Uttam Barman and Naznin Sultana. Clinical Evaluation of Recombinant Erythropoietin Biosimilar GBPD002 Compared with Eprex® in Adult Human. Archives of Clinical and Biomedical Research. 7 (2023): 447-462.

Table 3: Pharmacodynamics parameters after a single subcutaneous administration of the test and comparator drug products.

Parameters	Eprex® (n=41) Mean ± SD	CV	GBPD002 (n=40) Mean ± SD	CV	GMR (90% CI)[a]	p-value (ANOVA)
Reticulocyte (RET)						
T_{max} (h)[b]	108.48 ±2.70	-	127.2±2.81	-	-	-
ΔE_{max} (%)	91.94 ± 46.04	0.51	96.29 ± 40.88	0.42	1.047	0.604
Δ AUC (h.%)	642.29 ± 328.85	0.53	704.59 ± 115.0	0.16	1.097	0.976
Hemoglobin (HB)						
T_{max} (h)	118.56±3.61	-	86.4±1.20	-	-	-
ΔE_{max} (g/dL)	5.45 ± 3.32	0.61	4.90 ± 1.73	0.34	0.901	0.747
Δ AUC (h. g/dL)	37.9 ± 14.50	0.38	37.86 ± 11.60	0.31	0.999	0.859
Hematocrit (HCT)						
T_{max} (h)	122.4±3.62	-	123.36±3.84	-	-	-
ΔE_{max} (%)	4.57 ± 3.71	0.48	4.55 ± 2.64	0.58	0.999	0.970
Δ AUC (h.%)	95.57 ± 64.38	0.67	96.46 ±57.08	0.59	1.009	0.572
Red blood cell (RBC) count						
T_{max} (h)	126.24±3.45	-	76.32±3.38	-	-	-
ΔE_{max} (10^6/mm^3)	6.89 ± 2.69	0.52	6.07 ± 3.42	0.56	0.890	0.604
Δ AUC (h.10^6/mm^3)	43.67 ± 28.08	0.64	43.35 ± 30.54	0.70	0.993	0.976

N.B.: CV: coefficient of variation, SD: standard variation, CI: confidence interval, GMR: geometric mean ratio aGMR: (90% CI) of the test to the comparator epoetin alfa, b T_{max} (h): Mean (lowest-highest), $AUC_{0-144 h}$: area under the curve from time zero to the time of the last observation, AUC_{0-inf}: area under the curve extrapolated to infinity, C_{max}: maximum observed serum EPO concentration, CL/F: total clearance, MRT_{last}: mean residence time, t1/2: terminal half-life, aT_{max}: time of C_{max}; SD, standard deviation, aMean difference (90% CI) between the test and the comparator epoetin alfa, ANOVA: analysis of variance, ΔAUEC: area under the baseline-adjusted effect curve, ΔE_{max}: maximum effect change, bT_{max}: time of E_{max}.

Table 4: Reported adverse events during the course of the study

Parameters	Eprex®; (n=41)	GBPD002; (n=40)	Severity (mild/moderate/major)	Specific reactions related to the drug
General Weakness	12	13	mild	Non-specific to the drug
Headache	8	6	mild	Likely linked to the drug
Fever	3	2	mild	Non-specific to the drug
Sore throat	2	2	mild	Non-specific to the drug
Cough	2	2	mild	Non-specific to the drug
Nausea	1	0	mild	Non-specific to the drug
Abdominal pain	1	1	mild	Non-specific to the drug
Tinnitus	1	0	mild	Non-specific to the drug
Vertigo	3	0	mild	Non-specific to the drug
Back pain	4	1	mild	Non-specific to the drug
Neck pain	4	1	mild	Non-specific to the drug
Tingling	2	1	mild	Non-specific to the drug
Numbness	1	0	mild	Non-specific to the drug
Dyspepsia	1	2	mild	Non-specific to the drug
Cramping	1	2	mild	Non-specific to the drug
Itching	0	1	mild	Non-specific to the drug
Body-ache	0	2	mild	Non-specific to the drug
Chest discomfort	0	1	mild	Non-specific to the drug
Insomnia	0	1	mild	Non-specific to the drug
Focal weakness	0	1	mild	Non-specific to the drug
Diarrhea	2	1	mild	Non-specific to the drug
Total	48	40	0	0

Citation: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Mohammad Mohiuddin, Md. Abdur Rahim, Md. Helal Uddin, Samir Kumar, Md. Maksudur Rahman Khan, Md. Enamul Haq Sarker, Rony Roy, Sourav Chakraborty, Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Uttam Barman and Naznin Sultana. Clinical Evaluation of Recombinant Erythropoietin Biosimilar GBPD002 Compared with Eprex® in Adult Human. Archives of Clinical and Biomedical Research. 7 (2023): 447-462.

Discussion

The PK and PD parameters of two EPO formulations were evaluated and compared in this study. It has been shown that the pharmacological responses for PD markers (RET, HB, and RBC) were sex independent [26]. Therefore, we have included only male volunteers in our study. Yoon S et al., have recently performed the PK and PD study between a candidate EPO formulation with Eprex® in healthy male volunteers [16]. They have found comparable responses between the test and comparator for PK and PD parameters, and extrapolated their study findings sex-independently. In our study, we found that the PK and PD profiles and values of the test EPO (GBPD002) were similar to those of the comparator EPO (Eprex®). EPO is biologically removed from systemic circulation by attaching to its cognate receptor in the bone marrow [27]. The $t_{1/2}$ data in our study is in close proximity to other comparable studies [18-24], suggesting that the variables of the study were in coordination with other studies performed in different study sites and subjects. Previous studies reported that the PK of epoetin alfa may be influenced by the dynamic binding characteristics showing nonlinear disposition profiles [16]. In this study, a typical nonlinear elimination tendency was observed accordingly for both formulations. Other EPO receptor-binding drugs, such as epoetin beta and darbepoetin alfa, have also shown similar elimination patterns [28-30]. The RET count was shown to be highly predictive of erythropoiesis efficacy [31]. The dose-response association for the RET count with rHuEPO has been well established [32]. Further, the RET count has been recommended as the key PD marker in European and other recommendations for single-dose SC administration trials for EPO [33]. Therefore, RET count was determined as the key PD marker in this investigation. RET populations increased as the bone marrow resident hematopoietic cells responds to EPO. A RET number of fewer than 10,000/μL is considered to represent no or minimal regenerative response, 10,000–60,000/μL is a poor regenerative response, 60,000–200,000/μL is moderate response, and 200,000–500,000/μL is maximal regenerative response [34]. We have observed that the experimental formulation of EPO induced RET population significantly by day 3 and comparable with the reference product. RET population came down to the basal level on day 6, which is in accordance with the fact that the circulating RET population transformed to RBC within 2/3 days [35, 36]. This notion was supported by the findings that the HCT population went up on EPO administration on day 3, and the level was sustained after a slight dip. This data clearly suggested the classical effectiveness of both EPO preparations used in the study for RBC generation through the RET-induction pathway. Though minor and insignificant but a trend of very little higher response was observed for RET data for GBPD002 than the Eprex®. This observation can be assigned to the fact that the EPO function is dependent on resident time in the system rather than the higher level of concentration at a certain time point [37]. We also observed that the GBPD002 level remains a little higher in the system over the Eprex®, which is supportive to the notion. HB has been considered as another PD marker for EPO in several clinical studies. In our study, HB levels were found increased by 0.5 g/dl on day 3 after the SC administration of EPO and then declined to a normal level. Our result is in accordance with the findings of other studies where a similar level of raise in HB level was observed for SC-routed EPO administration [20, 23, 38, 39]. The RBC reading for our study also has shown similar results to the findings of Krzyzanski et al., and Sorgel et al., where the RBC level rises at a similar level on day 3 and then fell down to the basal level within 5/6 days [39, 40]. Yan et al., did not include RBC count in their study [41], therefore, a comparison of RBC response with their study was not possible. However, HB levels in all these studies and RBC levels in Krzyzanski et al., and Sorgel et al.'s, studies went up steadily after the administration of follow-on repeat doses. Considering very resembling trends for these PD markers for single-dose in duplicate segments, it can be expected that the responses for follow-on multiple doses for GBPD002 would be responding alike. Collectively, these results shown here demonstrated similar responses for PD markers for experimental EPO preparations, viz., GBPD002 and Eprex®. Counter-clockwise hysteresis was seen for both treatments when serum EPO concentrations and PD marker levels were time-matched. A counter-clockwise hysteresis refers to a time delay between the measured concentration and the PD reaction. The duration of exogenous EPO migrating from systemic circulation to its binding site in the bone marrow, as well as the delayed detection of the response, are possible sources of this indirect association [42, 43]. More specifically, the maturation of normoblasts into RET takes 5 – 7 days, and EPO plays a key part in this process [44].

No ADA was developed during the study period for either formulation. The main safety concern for EPO administration is the risk for the development of antibodies, which consequently develops epoetin-associated pure red cell aplasia (PRCA). EPO-associated PRCA was first reported in 1998 [45], and is characterized by severe anemia, low RET count, erythroblast absence, EPO nonresponse, and neutralizing antibodies [46 – 48]. Later the cause for antibody generation was attributed to leachates from the rubber stoppers and corrected by the application of Teflon coating to the rubber of the stopper [49]. The incidence of EPO-associated PRCA has significantly reduced thereafter [50,51]. In our study, we could not detect EPO-specific antibodies in any of the samples. Previous studies have also reported that ADA development is not common after subcutaneous injection of epoetin alfa [16], which has been suggested that non-specific immunoreactogenicity is unlikely

---

to happen for the EPO formulation under test. We have used a special polymer-coated leachate-free rubber stopper (West, Germany) for dose presentation in single-use PFS. Therefore, long-term risk for the generation of EPO-mediated PRCA from GBPD002 preparation is highly unlikely.

In conclusion, the PK and PD profiles of the test (GBPD002) epoetin alfa were identical to those of the comparator (Eprex®). The two medicines had similar levels of tolerability, including local toxicity and immunoreactogenicity profiles. The hysteretic connections between serum EPO levels and erythropoietic responses were similar for both products. Since, the two epoetin alfa medications have comparable pharmacokinetic, pharmacodynamics, and safety profiles therefore they can be considered biosimilar in clinical settings, and likely be administered interchangeably.

Funding

Author contributions

Conceptualization: Kakon Nag and Naznin Sultana; Test product manufacturing and evaluation: Samir Kumar, Md. Enamul Haq Sarker, Sourav Chakraborty, Md. Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Maksudur Rahman Khan, Rony Roy and Mohammad Mohiuddin; methodology and supervision of clinical trial: Kakon Nag, Naznin Sultana and Mohammad Mohiuddin; execution of clinical trial: Mamun Al Mahtab, Sitesh Chandra Bachar, Md. Abdur Rahim and Md. Helal Uddin; manuscript writing and editing: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Naznin Sultana, Mohammad Mohiuddin, Samir Kumar, and Md. Abdur Rahim; project administration: Kakon Nag and Md. Helal Uddin; All authors have read and agreed to the manuscript.

Declaration of competing interests

The authors declare that they have no competing interests.

Acknowledgements

Ethical statement

Ethics approval and consent to participate

The protocol for the study has got ethical clearance from the institutional review board (IRB; The Ethics Committee of Farabi General Hospital Ltd.) and approved by the Directorate General of Drug Administration (DGDA) of Bangladesh Ref. No. DGDA/CTP-1/06/2016/9916. The Clinical Research Organization (CRO Ltd.) conducted this investigation at Farabi General Hospital, Dhanmondi R/A, Dhaka 1209, in compliance with the principles of the Code of Ethics of the World Medical Association (Declaration of Helsinki) and the International Conference on Harmonization's Guideline for Good Clinical Practice (GCP).

The informed consent form (ICF), along with the protocol and methodology, were also approved by IRB and DGDA. Volunteers were given thorough information about the risks and benefits of the study and they have signed the ICF to affirm their willingness to voluntarily participate in the study. The trial protocol has been submitted and registered with the clinicaltrial.gov (ClinicalTrials.gov Identifier: NCT05585658) of the National Library of Medicine of the USA.

Data availability

The data that support the findings of this study are available within the article, and its Supplementary files, or are available from the corresponding author upon reasonable request.

Supplementary files

Supplementary information A.

References

1. Macdougall IC. Novel erythropoiesis-stimulating agents: A new era in anemia management. Clinical Journal of the American Society of Nephrology 3 (2008): 200-207.

2. Moore E, Bellomo R. Erythropoietin (EPO) in acute kidney injury. Ann Intensive Care 1 (2011): 3.

3. Bhoopalan SV, Huang LJ shen, Weiss MJ. Erythropoietin regulation of red blood cell production: from bench to bedside and back. F1000Res 9 (2020): 1153.

4. Hazin MAA. Anemia in chronic kidney disease. Rev Assoc Med Bras 66 (2020): 55-58.

5. Eschbach JW, Egrie JC, Downing MR, et al. correction of the anemia of end-stage renal disease with recombinant human erythropoietin. N Engl J Med 316 (1987): 73-78.

6. Eschbach JW. Recombinant human erythropoietin in anemic patients with end-stage renal disease: results of a phase iii multicenter clinical trial. Ann Intern Med 111 (1989): 992.

7. Lim VS. Recombinant human erythropoietin treatment in pre-dialysis patients: a double- blind placebo-controlled trial. Ann Intern Med 110 (1989): 108.

8. Ludwig H. Epoetin in cancer-related anaemia. Nephrology Dialysis Transplantation 14 (1999): 85-92.

9. Aapro M, Krendyukov A, Schiestl M, et al. epoetin biosimilars in the treatment of chemotherapy-induced anemia: 10 years' experience gained. BioDrugs 32 (2018): 129-135.

10. Biboulet P, Bringuier S, Smilevitch P, et al. preoperativeepoetin-α with intravenous or oral iron for major orthopedic surgery. Anesthesiology 129 (2018): 710-720.

11. Beiraghdar F, Panahi Y, Einollahi B, et al. Evaluation of a biosimilar recombinant alpha epoetin in the management of anemia in hemodialysis patients. Saudi Pharmaceutical Journal 23 (2015): 544-548.

12. Nag K, Haq Sarker MdE, Kumar S, et al. A cost- effective purification process for erythropoietin biosimilar through downstream process engineering [internet]. Bioengineering (2023).

13. Nag K, Islam MdJ, Rahman Khan MdM, et al. Development and qualification of a high-yield recombinant human Erythropoietin biosimilar [Internet]. Bioengineering (2023).

14. Approval of a protocol entitled ''A randomized, double-blinded, active controlled, single dosing, crossover clinical trial to investigate the pharmacokinetics, pharmacodynamics and safety of GBPD002 and Eprex® after subcutaneous administration in healthy adult volunteers'' (2021).

15. A Randomized, Double-blinded, Active Controlled Crossover Clinical Trial to Investigate PK, PD and Safety of GBPD002, ClinicalTrials.gov Identifier (2022).

16. Yoon S, Rhee S jin, Heo SJ, et al. Comparable pharmacokinetics and pharmacodynamics of two epoetin alfa formulations Eporon®; and Eprex®; following a single subcutaneous administration in healthy male volunteers 11 (2017): 3127-3135.

17. Schrag M, Mueller C, Oyoyo U, et al. Iron, zinc and copper in the Alzheimer's disease brain: A quantitative meta-analysis. Some insight on the influence of citation bias on scientific opinion. Progress in Neurobiology 94 (2011): 296-306.

18. Togawa A, Tanaka T, Nagashima S, et al. A comparison of the bioequivalence of two formulations of epoetin alfa after subcutaneous injection. Br J Clin Pharmacol 58 (2004): 269-276.

19. Grampp G, McElroy PL, Camblin G, et al. Structure-function relationships for recombinant erythropoietins: a case study from a proposed manufacturing change with implications for erythropoietin biosimilar study designs. Journal of Pharmaceutical Sciences 107 (2018): 1512-1520.

20. Ramakrishnan R, Cheung WK, Farrell F, et al. Pharmacokinetic and pharmacodynamic modeling of recombinant human erythropoietin after intravenous and subcutaneous dose administration in cynomolgus monkeys. J Pharmacol Exp Ther 306 (2003): 324-331.

21. Yoon S, Rhee S jin, Heo SJ, et al. Comparable pharmacokinetics and pharmacodynamics of two epoetin alfa formulations Eporon® and Eprex® following a single subcutaneous administration in healthy male volunteers 11 (2017): 3127-3135.

22. Macdougall IC. Optimizing the use of erythropoietic agents - pharmacokinetic and pharmacodynamic considerations. Nephrology Dialysis Transplantation 17 (2002): 66–70.

23. Yan X, Krzyzanski W. Quantitative assessment of minimal effective concentration of erythropoiesis-stimulating agents. CPT: Pharmacometrics & Systems Pharmacology 2 (2013): 62.

24. Kim TE, Kim KP, Kim BH, et al. Pharmacokinetic and pharmacodynamic comparison of two recombinant human erythropoietin formulations after single subcutaneous administration: An open-label, sequence-randomized, two-treatment crossover study in healthy Korean male volunteers. Clinical Therapeutics 32 (2010): 1968-1976.

25. Moradi Z, Maali A, Shad JS, et al. Updates on novel erythropoiesis-stimulating agents: clinical and molecular approach. Indian J Hematol Blood Transfus 36 (2020): 26-36.

26. Krzyzanski W, Jusko WJ, Wacholtz MC, et al. Pharmacokinetic and pharmacodynamic modeling of recombinant human erythropoietin after multiple subcutaneous doses in healthy subjects. European Journal of Pharmaceutical Sciences 26 (2005): 295-306.

27. Watowich SS. The erythropoietin receptor: molecular structure and hematopoietic signaling pathways. J Investig Med 59 (2011): 1067-1072.

28. Levesque B, Flynn M, Peters K, et al. P051 Phase 1a Safety and Pharmacokinetic Effects of GB004, a Novel Prolyl Hydroxylase Inhibitor and Potential Therapy for Inflammatory Bowel Disease. Am J Gastroenterol 114 (2019): 13.

29. Peletier LA, Gabrielsson J. Dynamics of target-mediated drug disposition: characteristic profiles and parameter identification. J Pharmacokinet Pharmacodyn 39 (2012): 429-4 51.

30. Halstenson CE, Macres M, Katz SA, et al. Comparative pharmacokinetics and pharmacodynamics of epoetin alfa and epoetin beta. Clin Pharmacol Ther 50 (1991): 702-712.

31. Han H, Lee J, Shin D, et al. Pharmacodynamics, pharmacokinetics, and tolerability of intravenous or subcutaneous gc1113, a novel erythropoiesis-stimulating agent. Clin Drug Investig 34 (2014): 373-382.

32. Davidkova S, Prestidge TD, Reed PW, et al. Comparison of reticulocyte hemoglobin equivalent with traditional markers of iron and erythropoiesis in pediatric dialysis. Pediatr Nephrol 31 (2016): 819-826.

33. Nguyen LM, Meaney CJ, Rao GG, et al. application of reticulocyte- based estimation of red blood cell lifespan in anemia management of end-stage renal disease patients. AAPS J 22 (2020): 40.

34. European Medicines Agency (EMA) Committee for Medicinal Products for Human Use. Guideline on non-clinical and clinical development of similar biological medicinal products containing recombinant erythropoietins (Revision) (2010).

35. Washington IM, Van Hoosier G. clinical biochemistry and hematology. in: the laboratory rabbit, guinea pig, hamster, and other rodents [internet]. Elsevier (2012).

36. Malleret B, Xu F, Mohandas N, et al. significant biochemical, biophysical and metabolic diversity in circulating human cord blood reticulocytes. Plos one 8 (2013): 76062.

37. Thomson-Luque R, Wang C, Ntumngia FB, et al. In-depth phenotypic characterization of reticulocyte maturation using mass cytometry. Blood Cells, Molecules, and Diseases 72 (2018): 22-33.

38. Besarab A, Flaharty KK, Erslev AJ, et al. Clinical pharmacology and economics of recombinant human erythropoietin in end-stage renal disease: the case for subcutaneous administration. Journal of the American Society of Nephrology 2 (1992): 1405-1416.

39. Sörgel F, Thyroff-Friesinger U, Vetter A, et al. bioequivalence of hx575 (recombinant human epoetin alfa) and a comparator epoetin alfa after multiple subcutaneous administrations. Pharmacology 83 (2009): 122-130.

40. Yan X, Krzyzanski W. Quantitative assessment of minimal effective concentration of erythropoiesis-stimulating agents. CPT: Pharmacometrics & Systems Pharmacology 2 (2013): 62.

41. Souillard A, Audran M, Bressolle F, et al. Pharmacokinetics and pharmacodynamics of recombinant human erythropoietin in athletes. Blood sampling and doping control. British Journal of Clinical Pharmacology 42 (1996): 355-364.

42. Lalka D. clinical pharmacokinetics: concepts and applications. Journal of Pharmaceutical Sciences 70 (1981): 973-974.

43. Tamilvanan S, Raja NL, Sa B, et al. Clinical concerns of immunogenicity produced at cellular levels by biopharmaceuticals following their parenteral administration into human body. Journal of Drug Targeting 18 (2010): 489-498.

44. Bleich HL, Boro ES, Krantz SB. Pure Red-Cell Aplasia. N Engl J Med 291 (1974) :345-350.

45. Prabhakar SS, Muhlfelder T. Antibodies to recombinant human erythropoietin causing pure red cell aplasia. Clin Nephrol 47 (1997): 331-335.

46. Peces R, de la Torre M, Alcázar R, et al. Antibodies against recombinant human erythropoietin in a patient with erythropoietin-resistant anemia. N Engl J Med 335 (1996): 523-524.

47. Casadevall N, Nataf J, Viron B, et al. Pure red-cell aplasia and antierythropoietin antibodies in patients treated with recombinant erythropoietin. N Engl J Med 346 (2002): 469-475.

48. Boven K, Stryker S, Knight J, et al. The increased incidence of pure red cell aplasia with an Eprex formulation in uncoated rubber stopper syringes. Kidney International 67 (2005): 2346-2353.

49. Woodburn KW, Fan Q, Winslow S, et al. Hematide is immunologically distinct from erythropoietin and corrects anemia induced by antierythropoietin antibodies in a rat pure red cell aplasia model. Experimental Hematology 35 (2007): 1201-1208.

50. Boven K, Knight J, Bader F, et al. Epoetin-associated pure red cell aplasia in patients with chronic kidney disease: solving the mystery. Nephrology Dialysis Transplantation 20 (2005): 33-40.

51. Macdougall IC, Casadevall N, Locatelli F, et al. Incidence of erythropoietin antibody-mediated pure red cell aplasia: the Prospective Immunogenicity Surveillance Registry (PRIMS). Nephrology Dialysis Transplantation 30 (2015): 451-460.

Supplementary information A

Supplementary Table 1: Pharmacokinetic parameter (serum EPO concentrations) after administration of Eprex®.

Volunteer ID	Day -1	Day 1	Day 3	Day 6	Day 8	Day 10	Day 14	Day 24	Day 48	Day 72	Day 96	Day 120	Day 144
1	6.820	41.935	68.550	23.130	93.800	48.700	32.760	20.245	20.245	13.520	26.500	25.055	7.113
2	4.913	5.390	145.315	18.800	12.085	15.920	1.112	9.690	25.055	16.345	11.708	10.170	4.436
3	11.262	25.535	99.650	79.250	125.500	87.000	49.670	20.245	12.560	13.520	11.708	10.960	8.480
4	11.259	33.725	140.970	14.000	32.280	7.775	20.245	21.205	13.040	14.436	11.709	10.205	3.960
5	11.255	31.315	48.700	14.480	148.220	27.460	21.205	8.255	19.760	12.534	9.130	7.865	7.110
6	11.260	50.799	69.550	100.689	32.280	13.040	36.140	38.070	36.140	29.919	13.960	10.170	7.104
7	11.258	50.803	91.400	202.534	131.205	106.500	98.255	79.690	51.594	29.924	12.534	8.009	7.107
8	11.257	3.484	61.300	100.689	35.655	16.880	5.865	24.090	21.587	19.920	11.140	8.960	6.645
9	11.261	4.436	140.485	30.350	27.460	23.610	23.130	1.112	11.590	11.918	11.712	7.725	7.111
10	11.258	50.802	77.800	16.400	47.735	53.550	28.425	27.741	7.300	0.170	11.705	11.758	7.108
11	11.256	50.799	82.650	16.400	100.150	78.250	107.950	60.800	11.590	9.921	4.913	9.210	21.685
12	1.112	150.799	1.112	10.170	13.520	12.560	1.585	9.690	11.591	9.917	2.534	29.870	2.534
13	2.059	63.250	53.550	105.000	53.050	27.945	28.425	15.440	21.685	9.923	11.714	7.775	23.130
14	11.262	3.009	35.655	50.650	38.555	48.700	34.690	154.500	14.960	32.760	11.708	7.300	14.000
15	11.262	50.801	27.460	14.480	52.100	148.000	56.450	49.185	2.534	55.500	11.708	4.436	5.390
16	15.920	50.799	20.245	144.150	36.140	18.800	22.165	14.960	6.820	11.125	11.711	15.440	3.009
17	11.262	103.799	3.484	6.820	39.035	27.460	28.905	19.280	29.390	3.484	11.708	0.170	7.113
18	11.259	3.009	88.177	0.000	14.480	1.585	46.285	131.800	10.170	9.210	11.712	11.761	0.170
19	11.255	20.725	44.835	74.850	38.070	45.315	39.520	24.570	11.585	9.923	11.705	6.820	7.113
20	11.260	50.796	71.950	19.280	84.100	63.700	43.385	42.900	11.592	9.915	11.713	11.758	18.320
21	11.262	0.170	24.090	76.800	41.935	47.250	40.970	5.865	11.593	10.645	11.712	3.960	2.560
22	11.261	1.634	96.120	150.600	113.685	108.390	95.760	69.990	37.890	9.918	11.707	11.762	7.109
23	11.264	12.939	38.070	92.550	50.670	52.155	33.870	12.320	20.625	9.915	11.716	21.810	7.112
24	10.238	219.450	220.500	288.650	245.700	207.600	118.095	38.265	11.586	9.919	11.719	11.757	7.106
25													
26	11.257	80.865	97.185	67.260	130.410	42.240	64.335	9.395	13.557	9.918	11.711	4.634	7.105
27	11.261	49.365	170.700	79.785	179.850	90.210	85.545	3.734	34.830	9.924	11.710	11.750	7.114
28	56.055	87.525	30.420	68.355	86.445	44.310	31.185	23.400	11.588	9.915	11.708	2.349	7.105
29	4.410	79.245	141.950	86.985	101.460	35.595	109.620	13.557	2.816	9.917	11.710	29.250	7.114
30	12.320	71.445	99.330	187.800	101.460	118.635	29.640	27.741	2.349	9.915	11.711	11.762	7.106
31	11.262	89.325	82.665	106.605	76.350	63.960	43.935	21.810	15.195	9.921	3.048	11.755	6.396
32	0.083	61.035	125.310	115.275	120.915	87.165	65.985	23.400	11.586	19.918	11.712	11.756	7.113
33	4.187	145.950	188.850	90.030	210.300	56.235	69.810	17.010	1.874	9.918	11.708	11.762	7.114
34	5.741	284.850	197.250	24.250	109.095	128.475	38.070	2.349	11.591	9.916	11.713	11.764	7.108
35	20.220	152.550	71.445	170.550	215.250	90.750	66.705	27.739	5.300	9.915	11.706	11.755	7.105
36	46.185	79.785	181.800	98.250	48.615	22.800	24.180	12.320	5.079	9.924	11.711	11.757	7.109
37	5.741	50.799	83.025	111.210	136.530	90.930	7.907	15.795	11.585	9.918	11.713	6.179	7.110
38	11.262	1.634	61.395	117.930	72.720	62.865	3.960	24.375	5.741	18.825	11.709	11.758	7.113
39	12.734	4.634	61.035	129.000	111.750	96.480	19.230	12.734	11.586	9.916	11.712	11.762	7.107
40	18.420	50.802	51.795	165.900	77.985	210.000	13.967	31.950	18.030	9.918	11.708	11.758	8.121
41	11.260	21.015	78.705	110.750	72.000	116.340	21.225	55.320	12.113	5.300	11.708	11.761	7.114
42	11.257	37.125	137.235	56.790	112.980	48.615	28.095	26.730	11.588	9.922	11.705	11.755	7.107
Avarage	12.13137	59.22314	88.09055	100.6888	87.11037	65.70122	42.39645	30.46985	15.09482	13.34585	11.46794	11.21878	7.650836
Median	11.261	55.9185	89.5725	102.4275	105.2775	75.5625	39.52	22.605	11.588	9.917989	11.7105	11.75739	7.109
Minimum	0.0825	0.1698	24.09	19.28	41.935	22.8	3.96	2.349	1.8735	5.2995	3.048	2.349	2.56
Max	56.055	284.85	220.5	288.65	245.7	210	118.095	69.99	37.89	19.91799	11.719	29.25	18.32
G. Mean	9.397438	33.39867	89.48555	94.42828	102.2319	74.11757	36.51354	18.00105	9.943903	10.27693	11.01555	10.37915	7.094133
SD	14.17578	65.04674	50.50358	41.03526	46.15055	43.82241	28.76527	12.04003	7.534426	3.331201	2.038038	5.286682	0.299719
CV	116.8522	109.8333	57.33144	40.75453	52.9794	66.69954	67.84829	39.51456	49.91398	24.96058	17.77162	47.1235	3.917463

Citation: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Mohammad Mohiuddin, Md. Abdur Rahim, Md. Helal Uddin, Samir Kumar, Md. Maksudur Rahman Khan, Md. Enamul Haq Sarker, Rony Roy, Sourav Chakraborty, Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Uttam Barman and Naznin Sultana. Clinical Evaluation of Recombinant Erythropoietin Biosimilar GBPD002 Compared with Eprex® in Adult Human. Archives of Clinical and Biomedical Research. 7 (2023): 447-462.

Supplementary Table 2: Pharmacokinetic parameter (serum EPO concentrations) after administration of the test drug GBPD002.

Volunteer ID	Day -1	Day 1	Day 3	Day 6	Day 8	Day 10	Day 14	Day 24	Day 48	Day 72	Day 96	Day 120	Day 144
1	13.859	37.695	76.170	73.455	120.750	117.930	47.310	51.600	14.786	21.120	17.466	13.213	10.520
2	13.862	40.077	101.300	107.029	101.628	82.275	69.331	42.059	15.972	21.118	17.467	13.140	10.515
3	13.855	49.365	114.750	78.525	117.930	93.255	68.535	42.064	15.970	24.117	18.473	11.209	10.518
4	13.860	9.816	25.755	60.105	57.165	133.905	47.310	42.057	10.865	16.815	17.470	17.213	5.524
5	13.864	7.692	50.115	146.755	63.420	55.680	59.565	27.705	37.125	19.816	17.473	15.960	8.448
6	41.868	77.805	90.750	45.435	39.030	67.995	25.350	15.966	11.118	7.473	6.210	3.515	3.515
7	13.855	40.077	101.300	107.029	101.628	82.275	12.527	42.064	15.966	14.115	9.473	6.211	6.515
8	6.830	12.734	80.505	144.210	104.655	87.425	47.895	36.560	24.880	19.183	17.473	14.213	10.520
9	61.770	48.435	54.570	73.995	129.360	46.935	85.005	42.064	31.575	11.120	8.473	6.208	6.515
10	0.633	40.545	40.725	82.845	63.045	43.185	13.146	26.535	52.155	11.121	17.466	14.211	2.519
11	13.862	17.820	124.440	131.280	115.110	44.505	47.260	40.300	25.350	20.119	17.467	16.209	6.518
12	18.420	17.010	33.105	73.635	68.715	50.670	31.575	17.415	18.760	11.121	9.473	9.214	4.516
13	13.861	40.077	101.300	107.029	105.195	82.275	41.075	42.063	15.966	11.118	7.473	4.206	5.517
14													
15	13.859	3.734	85.005	81.225	87.705	47.880	60.840	24.960	16.815	12.939	11.699	12.112	4.515
16	7.692	28.095	43.740	103.410	60.660	85.005	90.570	27.120	15.966	16.179	7.473	4.206	6.515
17	7.691	28.094	43.744	103.414	60.662	85.006	90.572	47.121	25.968	16.182	13.473	9.214	10.524
18	8.121	40.350	33.105	90.570	91.110	109.800	69.810	49.920	30.420	11.118	9.473	6.211	5.519
19													
20	7.907	40.077	75.075	72.180	49.185	102.885	60.300	42.059	15.966	1.874	17.473	4.208	10.523
21	13.860	27.514	31.575	22.994	56.610	35.990	17.415	10.451	25.755	26.402	17.473	4.206	10.521
22	13.856	27.510	31.569	22.995	56.613	35.985	17.411	10.448	25.763	26.396	17.466	14.211	10.518
23	13.856	14.000	120.600	69.550	122.050	218.550	124.500	59.350	25.971	11.118	17.467	5.390	8.320
24	19.760	26.980	165.100	20.245	28.425	29.870	21.205	26.980	15.970	10.170	17.473	9.210	3.750
25	1.112	31.315	53.550	63.700	78.750	47.735	50.150	39.035	15.965	11.605	9.690	4.913	5.390
26	13.864	18.320	22.645	107.029	101.628	28.425	69.329	42.055	15.968	19.760	13.484	8.255	8.090
27	22.645	6.820	86.050	36.140	78.250	67.100	54.500	35.655	23.610	21.123	17.470	12.560	8.240
28	2.534	54.000	126.900	173.900	136.900	104.450	89.950	78.750	48.220	21.119	17.467	11.213	7.840
29	44.835	16.880	19.760	107.029	50.650	82.275	85.050	49.185	35.175	11.121	14.913	8.730	5.390
30	13.857	40.077	71.450	26.500	134.250	104.050	93.900	88.000	36.345	19.125	17.473	4.205	7.775
31	13.861	40.077	70.000	148.700	40.970	44.835	28.905	17.840	0.640	11.117	17.469	4.214	6.820
32	12.560	99.077	101.300	28.905	26.980	58.850	130.850	19.280	15.966	11.121	7.471	5.865	4.360
33	13.859	95.077	113.300	127.029	88.800	82.275	33.520	23.200	15.971	11.119	7.100	6.205	5.905
34	6.345	100.077	101.300	117.650	121.400	54.000	46.765	31.950	23.610	24.200	17.000	4.210	6.820
35	7.300	59.077	95.400	107.029	74.436	62.275	65.750	42.061	25.966	28.150	25.055	13.960	10.484
36	13.863	91.245	112.150	284.500	194.550	84.100	69.329	42.064	15.973	13.800	7.473	3.484	4.515
37	11.125	117.077	145.400	137.585	95.300	66.600	87.500	91.400	15.920	12.085	17.465	3.995	7.300
38	6.820	86.077	101.100	158.300	243.800	180.250	146.000	60.800	5.865	13.040	17.471	3.960	3.085
39	14.675	59.405	84.531	99.988	91.613	78.079	64.875	43.951	20.696	14.786	14.957	87.241	9.727
40	13.855	40.077	48.700	56.500	43.865	42.900	70.500	40.485	7.300	21.685	17.473	34.436	7.300
41	13.861	149.280	161.350	157.400	59.900	62.200	59.700	27.250	8.730	18.320	17.469	9.690	8.730
42	13.858	184.077	123.050	153.550	152.850	132.300	73.400	62.250	15.970	11.118	7.471	9.690	3.515
Average	14.696	49.088	81.556	97.734	90.389	78.049	61.712	40.052	20.924	15.878	14.393	11.263	7.091
Median	13.857	40.077	95.400	107.029	78.750	66.600	65.750	42.055	15.971	13.800	17.467	6.205	7.300
Minimum	1.112	6.820	19.760	20.245	26.980	28.425	17.411	10.448	0.640	1.874	7.100	3.484	3.085
Max	44.835	184.077	165.100	284.500	243.800	218.550	146.000	91.400	48.220	28.150	25.055	87.241	10.523
G. Mean	11.076	46.457	77.582	78.481	79.213	68.070	58.577	36.914	16.023	14.383	14.506	7.836	6.752
SD	8.821	46.653	37.453	61.427	55.670	35.421	29.274	21.886	11.664	5.267	4.781	19.724	2.039
CV	60.026	95.038	45.924	62.852	61.589	45.383	47.437	54.645	55.745	33.171	33.221	175.122	28.755

Citation: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Mohammad Mohiuddin, Md. Abdur Rahim, Md. Helal Uddin, Samir Kumar, Md. Maksudur Rahman Khan, Md. Enamul Haq Sarker, Rony Roy, Sourav Chakraborty, Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Uttam Barman and Naznin Sultana. Clinical Evaluation of Recombinant Erythropoietin Biosimilar GBPD002 Compared with Eprex® in Adult Human. Archives of Clinical and Biomedical Research. 7 (2023): 447-462.

Supplementary Table 3: Reticulocyte count (%) after subcutaneous administration of the test or the comparator drug products.

	Eprex						GBPD002				
Volunteer ID	Day 0	Day 3	Day 6	Day 9	Day 14		Volunteer ID	Day 0	Day 3	Day 6	Day 9	Day 14
1	0.70	1.00	0.70	0.90	0.80		22	0.70	1.30	0.50	1.00	1.30
2	0.80	1.10	1.30	0.50	0.50		23	0.50	1.50	1.10	0.90	0.90
3	0.50	0.90	1.10	0.70	1.10		24	0.90	1.10	1.00	0.50	1.20
4	0.60	1.25	0.90	1.30	0.70		25	0.60	1.30	0.80	0.70	1.00
5	0.60	0.90	1.50	1.10	1.20		26	0.80	1.00	0.90	0.90	0.60
6	0.50	1.10	1.70	1.70	1.00		27	1.20	1.65	1.10	0.60	0.50
7	0.90	1.90	0.80	1.20	0.80		28	1.00	1.05	0.60	1.20	0.90
8	0.80	1.92	1.20	1.00	0.90		29	0.60	1.60	0.80	1.50	0.70
9	0.50	1.20	0.60	0.60	0.60		30	0.50	1.80	1.20	0.60	1.50
10	0.80	1.30	0.80	0.70	0.70		31	0.90	1.40	0.60	0.90	1.20
11	0.90	1.80	1.30	0.90	1.40		32	1.50	1.00	0.90	0.50	0.60
12	0.50	1.20	0.80	0.50	1.00		33	0.90	0.90	1.30	0.80	0.80
13	0.60	1.45	0.60	0.90	0.80		34	0.50	1.80	1.40	1.50	0.60
14	0.90	1.50	0.70	1.20	0.70		35	0.60	1.45	1.00	1.00	1.20
15	1.00	1.20	0.50	0.50	0.50		36	1.20	1.60	1.60	1.40	1.40
16	1.50	1.70	0.60	0.70	1.20		37	0.60	1.30	1.20	1.20	0.90
17	1.30	1.50	0.60	0.80	1.20		38	0.50	1.20	1.00	1.00	0.70
18	1.20	1.80	0.80	0.50	1.00		39	0.70	1.90	0.70	1.20	1.10
19	0.70	1.00	0.90	0.80	1.50		40	0.60	1.00	0.80	0.60	1.30
20	1.30	1.20	0.60	1.20	0.90		41	0.60	1.40	0.50	0.70	0.60
21	0.90	0.90	0.80	0.90	0.70		42	0.50	1.10	0.90	0.80	0.90
22	0.80	0.60	1.00	0.80	0.70		1	0.70	0.90	0.80	1.50	0.90
23	0.70	0.90	0.90	1.10	1.10		2	0.60	0.80	0.60	1.00	0.60
24	0.80	0.50	1.20	1.00	1.00		3	0.80	1.00	0.70	0.70	0.70
25							4	1.10	0.60	0.60	0.60	0.70
26	0.50	0.90	0.70	1.10	0.90		5	0.70	1.70	0.60	0.90	0.70
27	0.70	1.00	0.60	0.90	0.80		6	0.50	1.20	0.80	1.20	0.60
28	0.90	0.90	0.90	0.70	1.10		7	0.60	0.90	0.90	1.10	0.80
29	0.50	1.20	1.40	0.60	1.20		8	0.80	0.70	0.70	1.40	0.90
30	0.70	1.30	1.00	0.80	0.70		9	0.90	1.50	0.80	0.80	1.00
31	0.60	0.80	1.20	1.30	1.10		10	0.50	1.00	0.60	0.60	0.60
32	0.50	0.70	0.60	0.90	0.80		11	0.80	0.70	0.90	0.70	0.80
33	0.80	1.00	0.80	0.60	0.90		12	1.20	0.50	1.00	0.90	1.20
34	1.30	0.60	0.70	0.80	0.90		13	1.30	0.80	1.20	1.00	1.30
35	1.00	1.10	0.60	0.70	0.90		14					
36	0.90	0.90	0.80	1.50	0.90		15	0.50	0.60	0.80	0.70	0.90
37	0.60	0.80	0.60	0.90	0.80		16	0.70	0.90	0.90	0.60	0.80
38	0.90	1.10	0.80	0.70	0.60		17	0.50	1.00	0.70	1.70	0.90
39	0.60	1.00	0.70	0.80	0.90		18	0.70	0.80	0.60	0.60	0.90
40	0.90	0.80	0.80	1.00	0.70		19					
41	0.80	1.30	0.60	0.60	0.80		20	0.50	1.00	0.90	0.70	1.00
42	0.60	0.60	0.70	0.70	0.70		21	0.60	0.70	1.10	1.30	1.30
Avarage	0.80	1.12	0.86	0.88	0.90			0.75	1.14	0.88	0.94	0.91
S. Dev	0.35	0.25	0.28	0.27	0.22			0.36	0.25	0.25	0.32	0.26

Citation: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Mohammad Mohiuddin, Md. Abdur Rahim, Md. Helal Uddin, Samir Kumar, Md. Maksudur Rahman Khan, Md. Enamul Haq Sarker, Rony Roy, Sourav Chakraborty, Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Uttam Barman and Naznin Sultana. Clinical Evaluation of Recombinant Erythropoietin Biosimilar GBPD002 Compared with Eprex® in Adult Human. Archives of Clinical and Biomedical Research. 7 (2023): 447-462.

Supplementary Table 4: Hemoglobin (g/dl) after subcutaneous administration of the test or the comparator drug products.

	Eprex						GBPD002				
Volunteer ID	Day 0	Day 3	Day 6	Day 9	Day 14	Volunteer ID	Day 0	Day 3	Day 6	Day 9	Day 14
1	15.50	16.70	16.10	16.30	15.90	22	16.00	16.10	15.80	15.10	15.50
2	14.40	15.20	14.40	14.60	14.50	23	14.30	15.00	14.70	14.40	14.50
3	14.50	16.10	16.50	15.60	16.30	24	13.30	13.50	13.50	13.50	13.90
4	14.40	15.00	15.50	15.10	15.20	25	13.90	15.00	14.40	14.20	14.70
5	14.70	15.10	15.50	14.70	15.20	26	13.80	14.40	14.10	13.90	14.40
6	15.90	16.30	15.80	15.30	16.10	27	14.20	14.30	13.80	13.70	13.50
7	15.20	15.30	15.20	14.40	15.20	28	16.60	16.70	16.00	16.00	16.20
8	15.80	16.30	15.80	15.10	16.50	29	13.80	14.70	13.80	14.30	13.70
9	15.10	15.50	15.80	15.40	16.00	30	16.60	16.40	16.90	15.40	16.40
10	14.10	13.50	14.90	14.00	14.30	31	15.20	15.30	15.60	14.70	15.10
11	14.90	14.80	15.50	14.30	14.20	32	16.50	16.00	16.40	15.80	16.10
12	15.50	16.20	15.80	15.80	15.60	33	16.00	16.50	15.90	15.80	16.30
13	15.10	16.30	15.40	15.30	16.00	34	16.00	15.90	15.70	15.30	15.20
14	14.40	15.60	15.30	15.10	15.80	35	15.20	15.40	16.00	15.80	14.90
15	15.30	16.10	16.00	15.50	15.70	36	15.10	16.20	15.30	14.70	15.00
16	15.60	15.10	16.40	15.00	14.60	37	13.60	14.10	14.10	13.30	13.40
17	15.90	15.40	15.50	15.70	15.20	38	15.60	15.30	15.90	15.20	15.70
18	14.40	14.70	13.10	14.80	14.40	39	13.70	13.90	13.10	13.10	13.90
19	15.10	15.80	15.30	14.20	15.10	40	16.20	16.80	15.60	15.90	16.50
20	14.60	14.80	14.30	14.20	14.30	41	15.50	15.90	15.70	15.70	15.70
21	15.20	15.40	14.50	15.30	14.50	42	15.50	16.30	15.40	14.70	15.80
22	15.50	16.30	15.80	15.60	15.50	1	15.90	16.60	16.70	16.20	16.40
23	14.90	14.40	14.10	13.70	13.80	2	14.90	14.80	14.70	14.50	14.90
24	12.80	14.20	14.10	14.50	13.80	3	15.70	14.60	15.90	15.90	15.10
25						4	14.50	14.80	15.80	14.90	15.00
26	14.70	14.00	14.00	14.50	14.00	5	14.50	15.30	14.80	14.60	14.70
27	14.70	14.10	13.70	13.20	13.70	6	16.10	15.60	15.60	16.50	15.60
28	16.80	16.40	16.10	16.40	15.70	7	15.30	15.80	15.90	14.90	15.70
29	14.20	13.90	14.10	14.50	14.00	8	15.50	16.00	16.30	16.20	15.90
30	15.50	16.30	16.00	16.20	15.00	9	15.90	16.20	16.10	16.50	15.20
31	15.60	14.80	15.00	15.40	14.90	10	15.00	14.30	14.00	14.20	14.50
32	15.20	16.40	16.30	16.00	16.40	11	15.00	14.50	14.50	15.00	15.70
33	15.90	17.10	16.60	16.90	16.90	12	16.30	15.40	16.20	15.90	15.90
34	15.10	15.40	15.00	14.90	14.50	13	16.10	15.80	15.80	16.00	16.10
35	15.50	16.00	15.70	15.70	15.20	14					
36	14.80	15.50	14.40	15.10	15.20	15	15.00	15.50	16.00	14.70	16.00
37	13.00	13.10	13.40	12.70	13.90	16	15.40	15.60	15.00	15.40	15.10
38	16.00	15.50	15.90	15.00	16.00	17	14.90	15.00	15.10	15.20	15.00
39	13.50	13.10	13.30	13.00	13.40	18	14.10	13.40	14.10	14.20	13.60
40	16.30	15.90	16.20	15.80	16.30	19					
41	15.70	15.70	16.80	15.60	15.90	20	14.00	14.40	14.50	14.40	14.30
42	15.40	21.30	17.10	16.30	14.90	21	15.50	15.20	15.60	14.70	15.20
Avarage	15.04	15.48	15.27	15.04	15.11		15.16	15.31	15.26	15.01	15.16
S. Dev	0.79	1.31	0.98	0.90	0.88		0.90	0.86	0.91	0.86	0.84

Citation: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Mohammad Mohiuddin, Md. Abdur Rahim, Md. Helal Uddin, Samir Kumar, Md. Maksudur Rahman Khan, Md. Enamul Haq Sarker, Rony Roy, Sourav Chakraborty, Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Uttam Barman and Naznin Sultana. Clinical Evaluation of Recombinant Erythropoietin Biosimilar GBPD002 Compared with Eprex® in Adult Human. Archives of Clinical and Biomedical Research. 7 (2023): 447-462.

Supplementary Table 5: Hematocrit (%) after subcutaneous administration of the test or the comparator drug products.

	Eprex						GBPD002				
Volunteer ID	Day 0	Day 3	Day 6	Day 9	Day 14	Volunteer ID	Day 0	Day 3	Day 6	Day 9	Day 14
1	41.10	49.60	46.60	46.80	46.61	22	42.10	47.30	45.60	43.00	43.80
2	40.30	47.10	44.20	44.00	44.30	23	37.40	43.60	42.50	40.90	41.20
3	38.70	48.60	48.30	44.70	46.60	24	36.90	42.20	40.90	41.00	41.50
4	39.50	45.80	46.50	45.00	45.60	25	37.50	46.80	42.40	41.10	42.70
5	40.00	46.90	46.50	43.80	45.40	26	37.90	42.80	41.60	40.60	41.90
6	42.80	49.20	46.70	45.20	46.70	27	39.60	44.20	42.00	40.90	40.30
7	41.90	47.10	45.70	43.40	46.00	28	43.70	48.30	45.80	44.60	45.60
8	43.00	49.40	46.10	44.10	48.40	29	37.40	44.80	41.00	42.20	39.90
9	41.20	48.50	47.40	46.10	47.50	30	44.30	49.60	49.20	45.00	47.30
10	37.70	40.10	43.70	40.70	41.60	31	41.50	48.40	48.60	45.70	45.50
11	40.70	44.80	46.60	42.00	41.60	32	44.50	46.90	47.40	45.20	45.80
12	41.90	48.80	46.90	45.60	45.80	33	42.40	48.10	45.60	44.50	46.10
13	40.60	49.70	46.00	45.10	46.30	34	42.00	46.60	46.10	44.20	48.50
14	39.20	47.10	46.10	44.50	45.80	35	41.10	45.30	47.70	46.30	44.70
15	41.60	47.00	46.40	43.90	44.90	36	40.40	48.90	45.30	43.30	43.80
16	42.30	44.30	49.60	44.10	43.00	37	36.80	42.40	41.60	39.00	38.90
17	42.80	43.90	43.40	43.70	42.10	38	40.70	42.80	44.10	42.30	44.90
18	38.40	44.10	39.00	42.50	41.90	39	36.80	40.60	38.10	37.70	40.20
19	39.60	46.20	43.50	40.00	42.50	40	43.60	50.70	46.50	48.00	48.70
20	38.30	45.00	41.90	40.80	41.20	41	41.50	47.50	46.50	45.90	44.00
21	40.60	45.30	42.10	45.30	42.10	42	42.10	49.40	45.70	43.70	47.50
22	43.90	47.00	45.80	44.90	44.40	1	45.90	46.20	47.80	46.20	46.30
23	43.40	42.80	41.00	40.50	41.50	2	45.50	44.30	45.20	43.70	44.90
24	38.80	42.70	42.50	44.50	42.00	3	46.20	42.50	45.20	45.90	42.40
25						4	44.10	44.20	48.00	45.70	45.30
26	43.20	41.40	41.40	43.10	41.60	5	44.10	45.00	44.50	44.00	44.20
27	41.90	42.60	41.30	40.00	41.90	6	48.10	45.90	45.20	48.20	45.70
28	47.20	46.50	45.00	46.60	44.20	7	46.60	48.20	49.10	46.30	47.90
29	42.90	41.40	42.70	43.40	42.70	8	46.10	47.60	48.30	48.50	46.70
30	45.40	48.10	47.00	47.80	45.30	9	48.60	49.10	49.40	51.40	46.00
31	48.10	46.90	46.90	48.50	46.60	10	43.80	41.50	39.90	42.40	42.40
32	43.70	47.60	47.70	46.70	47.40	11	45.70	43.30	43.10	45.80	47.90
33	45.60	49.00	47.70	48.80	48.90	12	48.50	45.50	47.10	47.30	47.90
34	43.90	45.20	43.60	44.20	42.70	13	48.20	47.40	46.10	47.60	47.60
35	46.40	47.70	46.90	47.10	44.60	14					
36	42.50	45.50	42.80	45.30	44.30	15	43.10	44.50	46.10	42.90	46.40
37	37.80	38.70	39.10	37.30	41.00	16	45.90	47.00	44.50	46.00	45.10
38	44.60	44.00	46.10	43.20	45.70	17	42.50	42.80	42.70	42.90	43.50
39	38.60	37.90	38.50	37.60	38.80	18	41.50	39.70	41.90	42.10	39.70
40	48.60	47.90	48.80	48.20	49.40	19					
41	45.50	47.30	48.90	46.30	45.90	20	40.50	42.50	42.30	42.40	42.20
42	45.20	64.90	50.80	49.40	44.60	21	44.50	44.20	45.40	42.70	44.50
Avarage	42.18	46.19	45.07	44.26	44.38		42.74	45.47	44.90	44.18	44.49
S. Dev	2.82	4.14	2.92	2.77	2.43		3.13	2.69	2.70	2.69	2.60

Citation: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Mohammad Mohiuddin, Md. Abdur Rahim, Md. Helal Uddin, Samir Kumar, Md. Maksudur Rahman Khan, Md. Enamul Haq Sarker, Rony Roy, Sourav Chakraborty, Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Uttam Barman and Naznin Sultana. Clinical Evaluation of Recombinant Erythropoietin Biosimilar GBPD002 Compared with Eprex® in Adult Human. Archives of Clinical and Biomedical Research. 7 (2023): 447-462.

Supplementary Table 6: Red blood cell count (10^6/mm^3) after subcutaneous administration of the test or the comparator drug products.

	Eprex						GBPD002				
Volunteer ID	Day 0	Day 3	Day 6	Day 9	Day 14	Volunteer ID	Day 0	Day 3	Day 6	Day 9	Day 14
1	5.10	5.46	5.20	5.24	5.26	22	5.10	5.17	5.02	4.78	4.87
2	5.80	6.04	5.78	5.80	5.87	23	4.62	4.85	4.78	4.62	4.67
3	4.68	5.24	5.32	4.99	5.18	24	5.00	5.12	5.08	5.10	5.10
4	4.87	4.95	5.46	5.01	5.08	25	4.59	5.17	4.75	4.64	4.79
5	5.20	5.41	5.46	5.12	5.30	26	5.45	5.65	5.56	5.42	5.56
6	5.44	5.57	5.40	5.22	5.38	27	5.69	5.73	5.56	5.42	5.32
7	5.24	5.26	5.29	4.94	5.25	28	4.99	5.01	4.82	4.72	4.83
8	5.62	5.94	5.65	5.41	5.95	29	4.73	5.06	4.72	4.86	4.62
9	5.18	5.37	5.37	5.24	5.39	30	5.37	5.38	5.45	4.94	5.22
10	4.89	4.70	5.19	4.84	4.95	31	5.31	5.40	5.53	5.25	5.28
11	5.10	4.90	5.24	4.78	4.74	32	5.83	5.66	5.79	5.51	5.59
12	5.33	5.56	5.40	5.34	5.30	33	5.11	5.29	5.08	4.96	5.12
13	5.03	5.47	5.16	5.08	5.22	34	4.98	4.95	4.96	4.76	4.70
14	4.87	5.21	5.17	5.00	5.17	35	5.06	5.09	5.32	5.22	5.03
15	5.08	5.29	5.23	5.01	5.10	36	4.83	5.24	4.96	4.70	4.78
16	4.08	4.79	5.30	4.78	4.67	37	4.54	4.65	4.62	4.35	4.37
17	5.19	4.92	4.93	4.97	4.79	38	5.15	4.95	5.15	4.92	4.94
18	4.87	5.01	4.50	4.99	4.90	39	4.61	4.60	4.39	4.34	4.60
19	4.73	4.96	4.76	4.40	4.69	40	5.38	5.58	5.18	5.28	5.33
20	4.92	5.12	4.89	4.79	4.83	41	5.00	5.11	5.08	5.00	4.94
21	4.88	4.89	4.62	4.97	4.58	42	5.11	5.35	5.06	4.78	5.18
22	4.86	5.21	4.94	4.96	4.90	1	5.16	5.21	5.35	5.21	5.22
23	4.89	4.78	4.54	4.52	4.61	2	5.98	5.90	5.97	5.82	6.02
24	4.82	5.27	5.25	5.46	5.23	3	5.06	4.71	4.97	5.09	4.76
25						4	4.91	4.93	5.30	5.07	5.08
26	5.71	5.47	5.46	5.71	5.49	5	5.16	5.38	5.21	5.19	5.18
27	5.51	5.59	5.39	5.25	5.47	6	5.48	5.31	5.18	5.56	5.28
28	5.03	4.95	4.81	4.95	4.73	7	5.27	5.44	5.45	5.23	5.43
29	4.95	4.80	4.94	5.04	4.91	8	5.59	5.78	5.85	5.89	5.73
30	5.03	5.33	5.20	5.31	5.05	9	5.47	5.55	5.55	5.80	5.23
31	5.48	5.34	5.27	5.50	5.28	10	5.15	4.94	4.72	4.99	5.05
32	5.32	5.79	5.79	5.70	5.80	11	5.15	4.94	4.89	5.16	5.39
33	5.07	5.46	5.32	5.45	5.48	12	5.56	5.24	5.44	5.43	5.46
34	4.72	4.86	4.69	4.73	4.59	13	5.34	5.30	5.12	5.32	5.39
35	5.20	5.35	5.24	5.30	5.00	14					
36	4.67	4.98	4.67	4.93	4.86	15	4.89	5.04	5.16	4.84	5.23
37	4.26	4.37	4.32	4.22	4.58	16	4.93	5.08	4.80	4.96	4.87
38	5.10	5.05	5.17	4.96	5.24	17	4.83	4.83	4.84	4.90	4.91
39	4.40	4.34	4.33	4.30	4.40	18	4.71	4.55	4.77	4.81	4.56
40	5.34	5.37	5.30	5.28	5.43	19					
41	4.99	5.12	5.27	5.03	4.98	20	4.74	4.95	4.92	4.92	4.92
42	4.99	7.02	5.54	5.43	4.87	21	4.88	4.86	4.91	4.65	4.85
Avarage	5.04	5.23	5.14	5.07	5.09		5.12	5.17	5.13	5.06	5.09
S. Dev	0.35	0.45	0.35	0.35	0.36		0.34	0.32	0.35	0.36	0.34

Citation: Mamun Al Mahtab, Sitesh Chandra Bachar, Kakon Nag, Mohammad Mohiuddin, Md. Abdur Rahim, Md. Helal Uddin, Samir Kumar, Md. Maksudur Rahman Khan, Md. Enamul Haq Sarker, Rony Roy, Sourav Chakraborty, Bipul Kumar Biswas, Md. Emrul Hasan Bappi, Ratan Roy, Uttam Barman and Naznin Sultana. Clinical Evaluation of Recombinant Erythropoietin Biosimilar GBPD002 Compared with Eprex® in Adult Human. Archives of Clinical and Biomedical Research. 7 (2023): 447-462.