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Original Article

Efficacy, safety, and immunogenicity of recombinant insulin aspart (BioGenomics Limited) and NovoRapid® (Novo Nordisk) in adults with type 2 diabetes mellitus: a randomized, open-label, multicenter, phase-3 study

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Abstract

Background and purpose: To compare the efficacy, safety, and immunogenicity of recombinant insulin aspart 100 U/mL manufactured by BioGenomics Limited (BGL-ASP) with innovator NovoRapid® in type 2 diabetes mellitus patients (T2 DM).
Experimental approach: This was a multicenter, open-label, randomized, parallel-group study in T2 DM patients, on premix human insulin therapy ± oral anti-diabetics. Besides self-monitored plasma glucose, fasting and post-prandial plasma glucose (FPG and PPG) were tested at baseline, week 12, and week 24. Anti-insulin aspart antibodies measured immunogenicity at 12 and 24 weeks.
Findings/Results: 160 patients out of 320 patients randomly received BGL-ASP and the remaining patients received NovoRapid®. The changes in glycated hemoglobin (HbA_1c) from baseline to weeks 12 and 24 for the BGL-ASP group were -0.8 ± 0.83 and -0.8 ± 0.81, respectively, while for the NovoRapid®group was -0.8 ± 1.01 and -0.9 ± 0.89, respectively. Changes in FPG and PPG were comparable between the treatment groups after 12 weeks and 24 weeks. The incidence of detectable antibodies at baseline, weeks 12, and 24 were comparable between treatment groups. Eighteen (11.3%) patients in the BGL-ASP group and 23 (14.4%) in the NovoRapid®group reported adverse events.
Conclusion and implications: BGL-ASP and NovoRapid®were comparable and equally effective in lowering HbA1c, FPG, and PPG levels, with similar immunogenicity and safety profiles.

Keywords: Biosimilar; Immunogenicity; Insulin aspart; NovoRapid®; Type 2 diabetes mellitus.

INTRODUCTION

The Global Report on diabetes mellitus (DM) published by the World Health Organisation (WHO) in 2016 mentioned that about 422 million adults lived with DM in 2014, compared to 108 million in 1980. DM is a chronic illness that requires continuing medical care and patient self-management education to prevent risks of short-term and long-term complications (1). Since 1980 the worldwide prevalence of the adult population (age-standardized) of DM has increased from 4.7% to 8.5%. In comparison with high-income countries DM prevalence has increased more quickly in low- and middle-income nations during the past ten years. By raising the risks of cardiovascular and other illnesses, blood glucose levels that were higher than ideal were responsible for millions of fatalities.

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In low- and middle-income nations compared to high-income countries, a larger proportion of fatalities caused by high blood sugar or DM occur before the age of 70 years (2).

Recombinant insulin aspart 100 U/mL manufactured by BioGenomics Limited (BGL-ASP) is a rapid-acting analog of human insulin (HI) that swiftly lowers blood glucose. BGL-ASP is homologous with HI except for the amino acid proline substitution with aspartic acid at position 28 on the B-chain (3). Substitution of the proline residue at B28 with aspartic acid reduces the tendency of forming hexamers resulting in a faster rate of absorption, onset of action, and a shorter duration of action compared to HI (4). Hence, it has a quicker onset of action and more effective glucose-lowering action with superior control of post-prandial hyperglycemia and fewer nocturnal hypoglycaemic episodes than HI. Studies that noted an increase in cross-reactive insulin antibodies with aspart use showed a subsequent fall toward baseline values, without any indication of clinical relevance as there was no identified effect on efficacy or safety (5).

BGL-ASP has been developed as a biosimilar to Novo Nordisk's product NovoRapid®. As per various guidelines for developing biosimilars, it is important to demonstrate the similarity of the proposed biosimilar to the reference product in physicochemical and biological terms. Any observed differences were duly justified regarding their potential impact on safety and efficacy. The present study was conducted to compare the efficacy, safety, and immunogenicity of BGL-ASP (rDNA origin), I.P. injection) with NovoRapid® at week 12 and week 24.

MATERIALS AND METHODS

Study design and population

This was a multicenter (19 centers), open-label, randomized, parallel-group study in type 2 DM patients of either gender, age between 18-65 years, body mass index (BMI) $\geq$ 18 kg/m^2, and $\leq$ 40 kg/m^2, HbA_1c between 7.5%-10.0% (both inclusive), on premix human insulin therapy who may or may not be on oral antidiabetic drugs. The doses of oral antidiabetic drugs, if present, were to be stable for the previous three months before screening. Patients on the premix human insulin regimen were switched over to a basal-bolus regimen before screening where the bolus component was BGL-ASP or NovoRapid®, and the basal component was insulin glargine. The patients who met the inclusion criteria, willing to participate in the study were enrolled. The key exclusion criteria included the following: patients on insulin analogs (other than premix human insulin), known or suspected hypersensitivity to insulin or related product(s), patients who have participated in an interventional medical, surgical, or pharmaceutical study within 30 days before screening or are likely to simultaneously participate in another therapeutic clinical study, cardiovascular disease (in last 6 months from screening) or impaired liver function at the time of screening. The study was registered prospectively (CTRI/2019/04/018455). The study was conducted in compliance with ICH-GCP, New Drug and Clinical Trials Rules, 2019, and ICMR - Indian GCP Guidelines and prior approval from the EC was obtained. Before the beginning of the study, the investigator obtained the EC's approval for the written ICF, and all information was provided to the patients and/or their legal representatives as well as impartial witnesses. After screening procedures, each participant was assigned a unique randomization number, which identified the individual patient. Patients performed a 4-point / 7-point self-monitored plasma glucose (SMPG) and recorded the readings in a patient diary.

Dosing schedule for enrolled patients

The frequency of insulin glargine was once a day at bedtime and was not changed throughout the study. The dose of insulin glargine was adjusted based on investigators' discretion throughout the study. The frequency of BGL-ASP was once, twice, or thrice a day as per the patients' SMPG values. The dose titration was done using a weekly treat-to-target approach. The dose of insulin glargine was 20% lower than the basal component of the premix, and the dose of BGL-ASP was a unit-to-unit switchover from the bolus component of premix insulin (6).

Besides the SMPG, the fasting and post-prandial (2 h after a meal challenge) plasma glucose was tested in the laboratory at baseline, 12th week, and 24th weeks. The baseline values were tested on the day of randomization. At three time points of baseline, 12th week, and 24th week, the post-prandial sample was taken after a standard meal challenge. A standard meal was provided to the patients, comprising 60 g of Ensure Powder (Abbott, Indian) mixed with 250 mL of water. The ingredients per 100 g provide energy 435 Kcal, 20.1 g protein, 14.61 g fat, 0.73 g saturated fatty acids, 8.92 g monosaturated fatty acids, 1.14 g polysaturated fatty acids, 59.47 g carbohydrate and 5.19 g sugar. The total duration of the study was approximately 28 weeks from the day of screening (Fig. 1).

Selection and titration of doses

Patients performed SMPG and recorded the readings in a patient diary. At the end of each week, the site referred to the readings and calculated the average of the respective SMPGs, *e.g.* the average of three readings of pre-breakfast fasting SMPG was calculated, similarly the average of post-breakfast, the average of post-lunch and the average of post-dinner SMPG values were calculated. The basal insulin (insulin glargine) dose was titrated based on the fasting SMPG average, and the bolus insulin dose was titrated based on the post-prandial SMPG averages. The insulin doses for the subsequent week were titrated as per the recommended weekly titration.

Assessment parameters in study groups

Efficacy assessment was done by measuring and analyzing blood levels of HbA_1c, fasting plasma glucose (FPG), and post-prandial plasma glucose (PPG; after a standard meal challenge) at week 12 and week 24. Safety assessments included recording and analysis of adverse events, physical examination, vital signs, body weight, 12-lead electrocardiogram, and laboratory assessments.

Immunogenicity was assessed by measuring anti-insulin aspart antibodies at weeks 12 and 24. An enzyme-linked immunosorbent assay (ELISA) was developed in sequential steps for the detection of anti-insulin antibodies in the presence of aspart drug. Blood samples were collected before the start of the investigational product at the randomization visit (week 1), at the week 12 visit, and at the end of the treatment period *i.e.* the week 24 visit for testing immunogenicity. For each sampling visit, a blinded assessment was carried out to check anti-insulin antibody status (either positive or negative) and anti-insulin aspart titers.

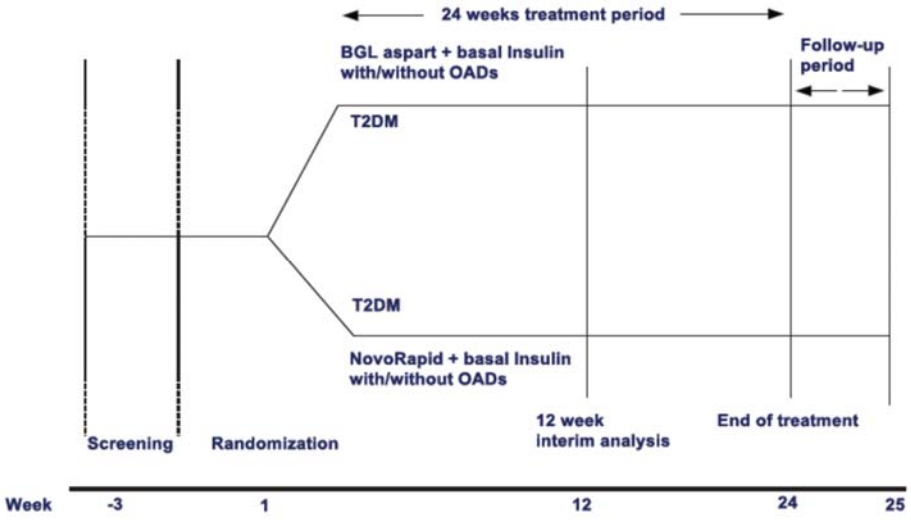

Fig. 1. Study flow chart describing key milestones for study. BGL, BioGenomics Limited; T2DM, type 2 diabetes mellitus; OADs, oral anti-diabetic drugs.

Sample size calculation and statistical analysis

Considering a non-inferiority margin of 0.4, standard deviation of 1.15, effect size of 0.33, power of 80%, and alpha error of 2.5%, the required sample size per group was noted to be 130 patients per group (260 patients for both study groups). Considering a 20% drop-out from the study over the follow-up period, the total sample size was considered 320 (160 per study group).

The descriptive statistics for continuous variables were presented with a number (n) of non-missing observations, mean, standard deviation, median, minimum, and maximum. For categorical data, the descriptive statistics were presented in the statistical report with the number of exposed patients and number (n) with the percentage of observations in various categories of the variables where the percentage was based on the exposed patients. Comparisons were made using the Chi-square/Fisher's exact test for categorical variables and paired t-test was used to compare within treatment arms while Student's t-test was used to compare within treatment arms for continuous measurement. Changes from baseline to week 12 and from baseline to week 24 in HbA_{1c} were also analyzed using a linear mixed model for repeated measurements. Efficacy analysis was done to test the study hypothesis using analysis of covariance (ANCOVA).

RESULTS

Patient disposition

A total of 565 patients were screened, of which 320 were randomized in the study. Of the 320 patients randomized, 160 patients received BGL-ASP and the remaining 160 patients received NovoRapid®. In total, 290 (90.6%) completed the study (Fig. 2). The duration of diabetes in years for both treatment arms was calculated from medical history data. The average diabetic history for the NovoRapid arm was 6.72 years and 8.29 years for the test drug BGL-ASP.

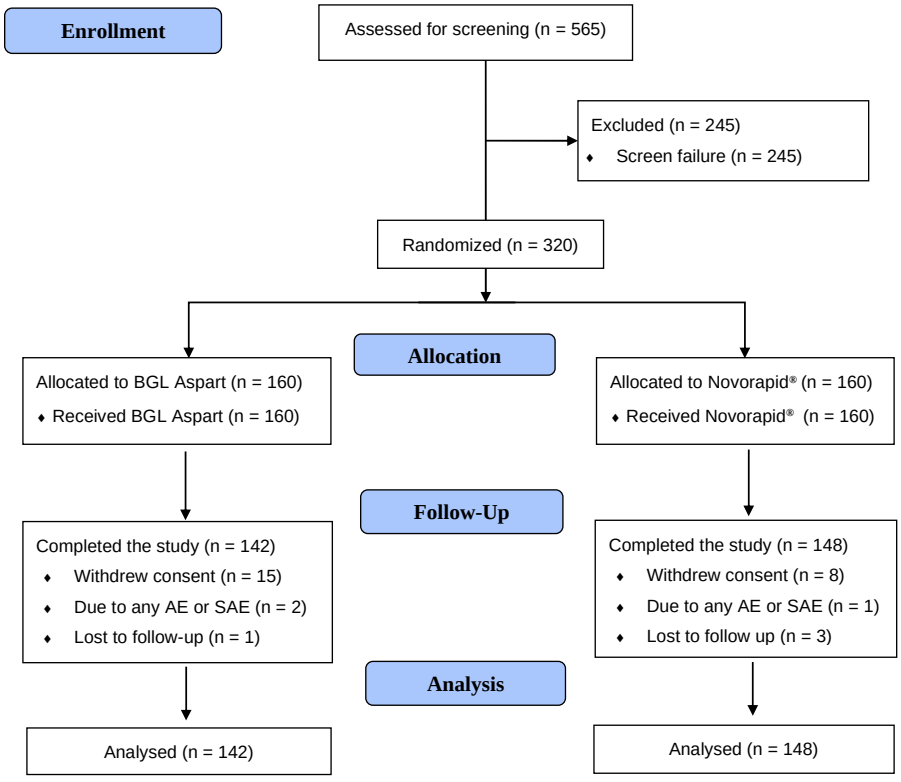

Fig. 2. Consort diagram. BGL-ASP, recombinant insulin aspart 100 U/mL manufactured by BioGenomics Limited; RMP, innovator NovoRapid®; AE, adverse event; SAE, serious adverse event.

Table 1. Demographic and anthropometric characteristics of patients at screening.

Parameters	BGL-ASP (N = 160)	NovoRapid® (N = 160)	Overall (N = 320)
Age (years)			
Mean	52.7	51.6	52.2
SD	8.11	9.23	8.69
Median	54.0	52.0	53.0
Range (min: max)	(29.0:65.0)	(30.0:65.0)	(29.0:65.0)
Gender, n (%)			
Male	90(56.3%)	79(49.4%)	169(52.8%)
Female	70(43.8%)	81(50.6%)	151(47.2%)
Height			
Mean	160.50	161.15	160.83
SD	8.46	9.56	9.02
Median	161.10	160.75	161.00
Range (min: max)	(138.0:183.0)	(132.0:185.0)	(132.0:185.0)
Weight			
Mean	71.19	69.39	70.29
SD	11.27	11.15	11.23
Median	69.25	68.20	69.00
Range (min: max)	(41.7:104.5)	(45.5:105.0)	(41.7:105.0)
Body mass index			
Mean	27.69	26.77	27.23
SD	4.26	4.05	4.17
Median	27.38	26.02	26.63
Range (min: max)	(18.7:39.3)	(19.1:39.4)	(18.7:39.4)
Waist circumference			
Mean	94.75	92.42	93.59
SD	12.03	11.07	11.60
Median	93.40	91.44	92.00
Range (min: max)	(71.1:138.0)	(67.0:122.0)	(67.0:138.0)

Demographic and anthropometric details in study groups

All 320 patients enrolled in the study were of Asian ethnicity. Out of 320 enrolled patients, 169 (52.8%) patients were males, of which 90 (56.3%) patients received BGL-ASP while 79 (49.4%) patients received NovoRapid®. The overall mean ± SD age (years) of the patient population was 52.2 ± 8.69 years (29-65 years); 52.7 ± 8.11 years was the mean age in the BGL-ASP group and 51.6 ± 9.23 years for NovoRapid® group. The anthropometric parameters like height, weight, BMI, and waist circumference were noted and were comparable between both groups (Table 1).

Medication characteristics at baseline

All 320 (100.0%) patients were on insulin, of which, a majority of 304 (95.0%) patients were taking human insulin and isophane combination as current medication. Hundred fifty one (94.4%) patients on human insulin were in the BGL-ASP group and 153 (95.6%) patients were in the NovoRapid® group. In the BGL-ASP group, 2 (1.3%) patients were taking insulin aspart; insulin aspart protamine (crystalline), and one patient each was taking insulin isophane bovine, insulin lispro, and isophane insulin, respectively as current insulin medication. In the NovoRapid® group, 2 (1.3%) patients were taking insulin isophane bovine. One patient each was taking insulin glargine and insulin lispro, respectively, as current insulin medication.

HbA_1c assessment at follow-up

The baseline amount of HbA1c was 8.8 ± 0.64 in the BGL-ASP group, while it was 8.7 ± 0.62 in the NovoRapid® group. The

changes in HbA1c from baseline to end of weeks 12 and 24 for the BGL-ASP group, were -0.8 ± 0.83 and -0.8 ± 0.81, respectively, while in the NovoRapid® group, it was -0.8 ± 1.01 and -0.9 ± 0.89, respectively (Table 2).

The estimate of HbA_1c in BGL-ASP and NovoRapid® groups at the end of week 12 were -0.72 and -0.88, respectively; the difference in the estimation of HbA1c was 0.16 ± 0.11 with 95% CI for difference estimate: -0.05:0.38) and *P*-value of 0.1416. The estimate of HbA_1c in the BGL-ASP and NovoRapid® groups at the end of week 24 were -0.78 and -0.96, respectively, the difference in the estimate of HbA_1c was 0.18 ± 0.10, with 95% CI for difference estimate -0.02:0.38 and *P*-value of 0.0783. By using ANCOVA analysis, the upper bound of the 95% CI for the difference in mean HbA_1c change from baseline between the treatment arms BGL-ASP and NovoRapid® was 0.38 at week 12 and week 24 achieving the primary endpoint.

At the end of week 12, 11 (7.4%) patients of the BGL-ASP group and 17 (11.2%) patients of the NovoRapid® group achieved $HbA_1c < 7\%$. At the end of week 24, 13 (8.7%) patients of the BGL-ASP group and 16 (10.5%) patients of the NovoRapid® group achieved $HbA_1c < 7\%$.

FPG assessment at follow-up

The changes in FPG from baseline to the end of week 12 and week 24 for the BGL-ASP group, were -7.1 ± 60.85 mg/dL and -18.8 ± 51.81 mg/dL, while for the NovoRapid® group was -15.8 ± 52.91 mg/dL and -14.9 ± 51.07 mg/dL. The results were comparable between the two treatment arms with a decrease in FPG from baseline to week 12 and baseline to week 24 (Table 3)

Using ANCOVA model analysis, the estimate of FPG in BGL-ASP and NovoRapid® groups at the end of week 12 were -8.54 and -14.45, and at the end of week 24 were -18.31 and -15.37, respectively. The difference in FPG estimate at the end of week 12 was 5.91 ± 4.7014.45 with 95% CI for difference estimate -3.35:15.18 and *P*-value of 0.2099, and at the end of week 24 was -2.94 ± 4.09, with 95% CI for difference estimate -11.01:5.1,1 and *P*-value 0.4722 which were comparable with no significant difference between groups.

Table 2. Summary of actual and change from baseline to the end of week 12 and week 24 in HbA1c parameter.

Statistics	Actual HbA_1c			Change in HbA_1c from baseline		
	BGL-ASP (N = 149)	**NovoRapid® (N = 152)**	**Overall (N = 301)**	**BGL-ASP (N = 149)**	**NovoRapid® (N = 152)**	**Overall (N = 301)**
			Screening visit			
N	149	152	301	NA	NA	NA
Mean	8.8	8.7	8.7	NA	NA	NA
SD	0.64	0.62	0.63	NA	NA	NA
95% CI	(8.71:8.92)	(8.57:8.77)	(8.67:8.82)	NA	NA	NA
***P*-value[1]**	NA	NA	NA	NA	NA	NA
***P*-value[2]**	NA	NA	**0.04**	NA	NA	NA
			Visit 5 (end of week 12)			
N	145	148	293	113	127	240
Mean	8.6	8.1	8.4	-0.8	-0.8	-0.8
SD	1.32	1.16	1.26	0.83	1.01	0.92
95% CI	(8.36:8.79)	(7.95:8.33)	(8.21:8.50)	(-0.93:-0.62)	(-1.02:-0.66)	(-0.93:-0.69)
***P*-value[1]**	NA	NA	NA	**< 0.001**	**< 0.001**	**< 0.001**
***P*-value[2]**	NA	NA	**0.003**	NA	NA	NA
			Visit 8 (end of week 24)			
N	144	148	292	96	109	205
Mean	8.4	8.2	8.3	-0.8	-0.9	-0.9
SD	1.16	1.18	1.18	0.81	0.89	0.85
95% CI	(8.21:8.59)	(7.97:8.36)	(8.14:8.41)	(-1.01:-0.68)	(-1.07:-0.74)	(-0.99:-0.76)
***P*-value[1]**	NA	NA	NA	**< 0.001**	**< 0.001**	**< 0.001**
***P*-value[2]**	NA	NA	0.09	NA	NA	NA

1, *P*-value was calculated using paired t-test within treatment arms; 2, *P*-value was calculated using Student's t-test between BGL-ASP and NovoRapid® groups; HbA1c, Glycated hemoglobin; BGL-ASP, recombinant insulin aspart 100 U/mL manufactured by BioGenomics Limited; CI, confidence interval; NA, not applicable; SD, standard deviation.

Table 3. Summary of actual and change from baseline to the end of week 12 and week 24 in FPG parameter.

Statistics	Actual FPG			Change in FPG		
	BGL-ASP (N = 149)	**NovoRapid® (N = 152)**	**Overall (N = 301)**	**BGL- ASP (N = 149)**	**NovoRapid® (N = 152)**	**Overall (N = 301)**
			Screening visit			
N	143	145	288	NA	NA	NA
Mean	144.2	145.1	144.6	NA	NA	NA
SD	49.2	46.9	47.9	NA	NA	NA
Median	131.0	132.0	131.5	NA	NA	NA
95% CI	(136.1:152.3)	(137.4:152.8)	(139.1:150.2)	NA	NA	NA
***P*-value[1]**	NA	NA	NA	NA	NA	NA
***P*-value[2]**	NA	NA	0.9	NA	NA	NA
			Visit 5 (end of week 12)			
N	132	138	270	132	138	270
Mean	134.9	129.3	132.0	-7.1	-15.8	-11.6
SD	41.2	36.6	39.0	60.8	52.91	57.0
Median	122.5	121.0	121.0	-8.5	-8.5	-8.5
95% CI	(127.8:142.0)	(123.1:135.5)	(127.4:136.7)	(-17.6:3.3)	(-24.70:-6.89)	(-18.4:-4.7)
***P*-value[1]**	NA	NA	NA	0.2	**0.0006**	**0.001**
***P*-value[2]**	NA	NA	0.2	NA	NA	NA
			Visit 8 (end of week 24)			
N	123	133	256	123	132	255
Mean	125.2	128.3	126.8	-18.8	-14.9	-16.8
SD	33.8	33.2	33.5	51.8	51.1	51.4
Median	115.0	120.0	118.0	-19.0	-15.5	-16.0
95% CI	(119.2:131.3)	(122.6:134.0)	(122.7:131.0)	(-28.1:-9.6)	(-23.7:-6.1)	(-23.1:-10.5)
***P*-value[1]**	NA	NA	NA	**< 0.001**	**0.001**	**< 0.001**
***P*-value[2]**	NA	NA	0.5	NA	NA	NA

1, *P*-value was calculated using paired t-test within treatment arms; 2, *P*-value was calculated using Student's t-test between BGL-ASP and NovoRapid®; FPG, fasting plasma glucose; BGL-ASP, recombinant insulin aspart 100 U/mL manufactured by BioGenomics Limited; CI, confidence interval; NA, not applicable; SD, standard deviation.

PPG assessment at follow-up

The changes in PPG from baseline to the end of week 12 and end of week 24 for the BGL-ASP group, were -54.5 ± 54.66 mg/dL and -54.5 ± 44.83 mg/dL, while for NovoRapid® was -49.9 ± 42.46 mg/dL and -48.0 ± 43.56 mg/dL. (Table 4).

Using ANCOVA model analysis, the estimate of PPG in BGL-ASP and NovoRapid® groups at the end of week 12 were -51.01 and -52.82, and at the end of week 24 were -52.36 and -49.90, respectively. The difference in the estimate of PPG at the end of week 12 was 1.81 ± 6.98 with 95% CI for difference estimate: -11.98:15.61 (*P*-value: 0.7952) and at the end of week, 24 was -2.45 ± 6.80, with 95% CI for difference estimate: -15.93:11.01 (*P*-value: 0.7185) which were not significantly different between the groups.

Immunogenicity outcomes

Nineteen patients of the BGL-ASP group were positive for anti-insulin aspart antibody at baseline; of these, 9 (47.4%) patients at week 12 and 5 (26.3%) patients at week 24 were positive. Of the 141 patients of the BGL-ASP group who were negative for the antibody at baseline, 5 (3.5%) patients at week 12 and 4 (2.8%) patients at week 24 were positive. In the NovoRapid® group, 16 patients were positive for anti-insulin aspart antibody at baseline of these, 4 (25%) patients at week 12 and 2 (12.5%) patients at week 24 were positive. Of the 143 patients of the NovoRapid® group who were negative for the antibody at baseline, 6 (4.2%) patients at week 12 and 10 (7%) patients at week 24 were positive. The incidence of detectable antibodies at baseline, week 12, and week 24 showed comparable findings between the treatment groups ($P > 0.05$).

Table 4. Summary of actual and change from baseline to end of week 12 and end of week 24 in PPG parameter.

Statistics	Actual PPG			Change in PPG		
	BGL-ASP (N = 149)	**NovoRapid® (N = 152)**	**Overall (N = 301)**	**BGL-ASP (N = 149)**	**NovoRapid® (N = 152)**	**Overall (N = 301)**
Screening visit						
N	138	140	278	NA	NA	NA
Mean	211.8	214.1	212.9	NA	NA	NA
SD	58.4	55.5	56.8	NA	NA	NA
Median	221.0	228.0	226.5	NA	NA	NA
95% CI	(202.0:221.6)	(204.8:223.4)	(206.2:219.6)	NA	NA	NA
***P*-value[1]**	NA	NA	NA	NA	NA	NA
***P*-value[2]**	NA	NA	0.73	NA	NA	NA
Visit 5 (end of week 12)						
N	134	135	269	73	87	160
Mean	219.7	202.9	211.3	-54.5	-49.9	-52.0
SD	66.6	65.7	66.54	54.6	42.5	48.3
Median	219.5	208.0	215.0	-34.0	-40.0	-36.5
95% CI	(208.3:231.1)	(191.7:214.0)	(203.3:219.2)	(-67.2:-41.7)	(-59.0:-40.9)	(-59.5:-44.5)
***P*-value[1]**	NA	NA	NA	**< 0.001**	**< 0.001**	**< 0.001**
***P*-value[2]**	NA	NA	**0.03**	NA	NA	NA
Visit 8 (end of week 24)						
N	131	137	268	59	67	126
Mean	212.9	205.1	208.9	-54.5	-48.0	-51.1
SD	73.8	64.7	69.3	44.8	43.6	44.1
Median	203.0	198.0	199.5	-37.0	-33.0	-34.5
95% CI	(200.2:225.7)	(194.2:216.1)	(200.6:217.3)	(-66.2:-42.8)	(-58.6:-37.4)	(-58.8:-43.3)
***P*-value[1]**	NA	NA	NA	**< 0.001**	**< 0.001**	**< 0.001**
***P*-value[2]**	NA	NA	0.35	NA	NA	NA

1, *P*-value was calculated using paired t-test within treatment arms; 2, *P*-value was calculated using Student's t-test between BGL-ASP and NovoRapid®; PPG, post-prandial plasma glucose; BGL-ASP, recombinant insulin aspart 100 U/mL manufactured by BioGenomics Limited; CI, confidence interval; NA, not applicable; SD, standard deviation.

The neutralizing capacity of antibodies is measured using *in-vitro* biological assays to check the function of the drug. The potential neutralizing capacity of anti-insulin aspart antibodies was analyzed using a validated glucose uptake assay. Patient serum samples collected at the end of the 6-month treatment period were analyzed for neutralizing antibody assay.

Twenty-one patients (9 (5.6%) patients of the BGL-ASP group and 12 (7.5%) patients of the NovoRapid® group) reported positive results for the incidence of detectable antibodies. However, the immunogenic response shown by these patients was low. The assay showed glucose uptake in all the confirmed positive serum samples, indicating the absence of neutralizing antibodies. The average glucose consumption of patients who received NovoRapid® as treatment was in the range of 80.34 to 83.48% whereas, that of patients who received BGL-ASP was in the range of 80.27 to 82.63%.

Safety analysis

Of an overall 320 patients enrolled in the study, 41 (12.8%) patients experienced 61 treatment-emergent adverse events (TEAEs); in the BGL-ASP group, 18 (11.3%) patients reported 24 events, whereas in the NovoRapid® group, 23 (14.4%) patients reported 37 events. One event reported in the NovoRapid®group was a serious adverse event (SAE), all other events were non-serious. Of an overall 61, 32 TEAEs were hypoglycaemic events out of which the majority were reported in the NovoRapid® group. In the BGL-ASP group, 9 (5.6%) patients reported 13 events, whereas in the NovoRapid® group, 11 (6.9%) patients reported 19 events. Table 5 shows the summary of TEAEs in the treatment group.

Table 5. Summary of treatment-emergent adverse events by treatment group.

Parameter statistics	BGL-ASP (N = 160)	NovoRapid® (N = 160)	Overall (N = 320)
	N, (% of N), [Total number of events]		
Total	18 (11.3%) [24]	23 (14.4%) [37]	41 (12.8%) [61]
Is it a serious adverse event?			
Yes	0 (0.0%) [0]	1 (0.6%) [1]	1 (0.3%) [1]
No	18 (11.3%) [24]	22 (13.8%) [36]	40 (12.5%) [60]
Severity grade			
Grade 1	17 (10.6%) [23]	19 (11.9%) [31]	36 (11.3%) [54]
Grade 2	1 (0.6%) [1]	3 (1.9%) [3]	4 (1.3%) [4]
Grade 3	0 (0.0%) [0]	1 (0.6%) [2]	1 (0.3%) [2]
Grade 4	0 (0.0%) [0]	1 (0.6%) [1]	1 (0.3%) [1]
Causality to study medication			
Certain	5 (3.1%) [6]	10 (6.3%) [19]	15 (4.7%) [25]
Probable	2 (1.3%) [2]	2 (1.3%) [4]	4 (1.3%) [6]
Possible	4 (2.5%) [8]	3 (1.9%) [5]	7 (2.2%) [13]
Unlikely	8 (5.0%) [8]	8 (5.0%) [9]	16 (5.0%) [17]
The withdrawn patient due to an adverse event			
Yes	1 (0.6%) [1]	1 (0.6%) [1]	2 (0.6%) [2]
No	17 (10.6%) [23]	22 (13.8%) [36]	39 (12.2%) [59]
Expectedness			
Expected	10 (6.3%) [14]	11 (6.9%) [19]	21 (6.6%) [33]
Unexpected	9 (5.6%) [10]	12 (7.5%) [18]	21 (6.6%) [28]
Action taken with study medication			
None	12 (7.5%) [15]	12 (7.5%) [20]	24 (7.5%) [35]
Product withdrawn	0 (0.0%) [0]	1 (0.6%) [1]	1 (0.3%) [1]
Dose reduced	5 (3.1%) [7]	10 (6.3%) [16]	15 (4.7%) [23]
Dose increased	1 (0.6%) [1]	0 (0.0%) [0]	1 (0.3%) [1]
Other (Specify)	1 (0.6%) [1]	0 (0.0%) [0]	1 (0.3%) [1]
Action taken to manage the event			
None	5 (3.1%) [5]	3 (1.9%) [5]	8 (2.5%) [10]
Drug therapy started	5 (3.1%) [5]	9 (5.6%) [10]	14 (4.4%) [15]
Test performed	1 (0.6%) [1]	0 (0.0%) [0]	1 (0.3%) [1]
Other (specify)	8 (5.0%) [13]	12 (7.5%) [22]	20 (6.3%) [35]
Outcome			
Recovered	18 (11.3%) [24]	23 (14.4%) [37]	41 (12.8%) [61]

BGL-ASP, recombinant insulin aspart 100 U/mL manufactured by BioGenomics Limited.

DISCUSSION

A multistep method is required to prove biosimilarity, which is demanding but difficult at the same time (7). To create data in support of biosimilarity and assess any remaining doubt, a step-by-step, totality-of-evidence-based strategy must be utilized, according to European Medicines Agency (EMA) and United States Food and Drug Administration (FDA) advice on the development and approval of biosimilars (8,9). The FDA criteria to produce biosimilar insulin have changed because of significant experience and information on the availability of insulin and biosimilars (10). In the European Union, safety investigations should typically be carried out with an emphasis on immunogenicity. Studies on immunogenicity research are required before biosimilars like insulin may be approved since the immunogenicity metric may be connected to process- and product-related contaminants. The objective of the current study was to compare the immunogenicity characteristics of the treatment groups (11) and the HbA1C, FPG, and PPG between the two study groups (12). To guarantee adequate patient exposure to identify variations in immunogenicity, effectiveness, and safety factors, the study's design including its main and secondary endpoints, population selection, and treatment duration complied with US-FDA scientific recommendations.

According to the ANCOVA analysis, the study's findings for the BGL-ASP and NovoRapid® groups were comparable, with

reductions in HbA1c from baseline to week 12 and week 24. The mean change in FPG and PPG from baseline to end of week 12 and from baseline to end of week 24 were tested as the secondary efficacy endpoint, and the results were comparable between the two treatment arms with no significant difference. At the end of week 12 and week 24, the percentage of patients who met the A1c target (< 7%) was comparable between the two treated groups. These findings showed the comparability of BGL-ASP with NovoRapid® in terms of efficacy parameters.

At baseline, week 12, and week 24, the incidence of detectable antibodies was examined, and the findings implied that both treatment groups were comparable. This demonstrated that the tested biosimilar agent and the innovator had comparable immunogenic potential. Type 2 DM population represents most patients with diabetes, the generation of data is therefore considered appropriate to evaluate outcomes in a wide group of the population with diabetes. The published research by Garg *et al.* showed a comparable immunogenic response between the two populations with diabetes (type 1 and type 2 DM) during the primary 24-week treatment phase for the therapy-induced anti-insulin aspart antibody (13). Therefore, even though type 1 DM patients were not included in the study, the findings from it may be regarded as typical of the whole DM community.

While 23 (14.4%) patients in the NovoRapid® group had TEAEs, just 18 (11.3%) patients in the BGL-ASP group experienced the same. In the BGL-ASP group, in 9 (5.6%) patients 13 hypoglycaemic episodes were reported, while in the NovoRapid® group, 11 (6.9%) patients reported 19 hypoglycaemic events. These results were consistent with those of previous studies that compared originator insulin analogs with biosimilars (14). In the present investigation, the incidence of hypoglycemia episodes was comparable between the two groups and was significantly lower than the 17% reported for Ref-InsAsp-US in a randomized controlled trial (15). However, when contrasted with research that compared biosimilar insulin aspart (MYL1601D) with originator insulin aspart (Novolog®), the proportion of TEAE occurrences was reduced (16).

Limitations of the current study which are worth mentioning include phase 3 studies conducted to examine the efficacy of diabetes agents relying, by design, on short-term assessment of physiological endpoints, such as A_1C levels, which are considered predictive of improved medical outcomes when followed over time. In addition, phase 3 studies are designed to observe safety in relatively modest numbers of individuals over just a 6- to 12-month interval. Hence, both long-term benefits and risks are only partly assessed. Also, this study was conducted in only type 2 diabetes patients. All the above limitations can be overcome in the phase 4 study.

CONCLUSION

Twelve-week and 24-week administrations of BGL-ASP and NovoRapid® were comparable and equally effective in lowering the HbA_1c, FPG, and PPG levels of type 2 DM patients, meeting primary and secondary endpoints of the study. Furthermore, no relevant differences in anti-insulin aspart antibodies were observed between the two treatment groups. The safety profile of BGL-ASP was comparable to NovoRapid®. BGL-ASP has proven to be non-inferior to NovoRapid® in treating type 2 DM patients and can be a recommended treatment option.

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Conflict of interest statement

The authors declared no conflict of interest in this study. All co-authors have seen and agreed with the content of the manuscript.

Authors' contributions

S.A. Dongre and G.A. Kulkarni contributed equally to this study. S.A. Dongre,

G.A. Kulkarni, and N. Sonar contributed to writing of the manuscript; S.A. Dongre, G.A. Kulkarni, A. Mishra, and R.B. Deshmane contributed to immunogenicity study; K. Yashi and S. Kadoo contributed to the review of overall clinical aspect of the study; D. Thapa, N. Ghade, S.M. Sonar, and A.R. Krishnan contributed to conception, design, and development of the molecule, review of important intellectual content, and final approval to be published. All the authors have reviewed and approved the finalized article.

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