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ORIGINAL ARTICLE

Greater early postprandial suppression of endogenous glucose production and higher initial glucose disappearance is achieved with fast-acting insulin aspart compared with insulin aspart

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Aim: To investigate the mechanisms behind the lower postprandial glucose (PPG) concentrations achieved with fast-acting insulin aspart (faster aspart) than with insulin aspart (IAsp).

Materials and methods: In a randomized, double-blind, crossover trial, 41 people with type 1 diabetes received identical subcutaneous single faster aspart and IAsp doses (individualized for each participant), together with a standardized mixed meal (including 75 g carbohydrate labelled with [1-^{13}C] glucose). PPG turnover was determined by the triple-tracer meal method using continuous, variable [6-^{3}H] glucose and [6,6-$^{2}H_2$] glucose infusion.

Results: Insulin exposure within the first hour was 32% greater with faster aspart than with IAsp (treatment ratio faster aspart/IAsp 1.32 [95% confidence interval {CI} 1.18;1.48]; $P < .001$), leading to a 0.59-mmol/L non-significantly smaller PPG increment at 1 hour (ΔPG_{1h}; treatment difference faster aspart–IAsp −0.59 mmol/L [95% CI −1.19; 0.01]; $P = .055$). The trend towards reduced ΔPG_{1h} with faster aspart was attributable to 12% greater suppression of endogenous glucose production (EGP; treatment ratio 1.12 [95% CI 1.01; 1.25]; $P = .040$) and 23% higher glucose disappearance (1.23 [95% CI 1.05; 1.45]; $P = .012$) with faster aspart than with IAsp during the first hour. Suppression of free fatty acid levels during the first hour was 36% greater for faster aspart than for IAsp (1.36 [95% CI 1.01;1.88]; $P = .042$).

Conclusions: The trend towards improved PPG control with faster aspart vs IAsp in this study was attributable to both greater early suppression of EGP and stimulation of glucose disappearance.

KEYWORDS

glucose metabolism, insulin therapy, pharmacodynamics, pharmacokinetics, type 1 diabetes, type 2 diabetes

1 | INTRODUCTION

Postprandial glucose (PPG) levels are important in determining overall glycaemic control[1,2]; therefore, reduction of postprandial hyperglycaemia is key to achieving the recommended glycaemic profile over 24 hours in people with diabetes.[3] Fast-acting insulin aspart (faster aspart) is an ultra-fast-acting mealtime insulin developed to provide better PPG control than that achieved with current rapid-acting insulin analogues. Faster aspart is insulin aspart (IAsp) in a new formulation, containing two well-known additional excipients, L-arginine and niacinamide, and resulting in a stable formulation with accelerated initial absorption after subcutaneous administration.[4,5] Faster aspart has twice-as-fast onset of appearance, 2-fold higher early exposure and 74% greater early glucose-lowering effect

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compared with IAsp, leading to reduced PPG concentration with faster aspart.[4,6–9]

The PPG concentration is determined by the rate of appearance of ingested glucose (ie, glucose absorbed from the meal), the rate of endogenous glucose production (EGP; ie, hepatic glucose output) and the rate of glucose disappearance (glucose R_d; primarily muscle glucose uptake).[10] Physiological insulin secretion after a meal in healthy people leads to rapid and profound suppression of EGP combined with increased glucose R_d, thereby controlling the temporary rise in glucose concentration.[11] In contrast, people with type 1 diabetes (T1D) lack endogenous insulin, and in people with type 2 diabetes (T2D) postprandial suppression of EGP is slower and the glucose R_d is lower for given circulating glucose and insulin concentrations, with both factors contributing to postprandial hyperglycaemia.[12,13] For mealtime insulins, to ensure the best control of PPG, it would be desirable to replicate the insulin profile in the healthy state, thus ensuring rapid and early stimulation of glucose uptake as well as suppression of EGP.

With the accelerated pharmacokinetic profile of faster aspart, closer to physiological prandial insulin delivery, we hypothesized that the reduced postprandial hyperglycaemia achieved with faster aspart resulted not only from enhanced early stimulation of glucose uptake, but also from greater early inhibition of EGP. To test this hypothesis, in the present study, we investigated the mechanisms behind the improved PPG control with faster aspart vs IAsp, using state-of-the-art triple-tracer methodology to assess PPG turnover.[10,14]

2 | MATERIALS AND METHODS

2.1 | Study design

This was a randomized, single-centre (Department of Internal Medicine, Division of Endocrinology and Metabolism, Medical University of Graz, Austria), double-blind, two-period, crossover trial in people with T1D. The trial protocol was reviewed and approved by the Independent Ethics Committee of the Medical University of Graz, and by the appropriate health authorities according to local regulations. The trial was performed in accordance with the Declaration of Helsinki and Good Clinical Practice. Written informed consent was provided by all participants before initiation of any trial-related activities. The trial was registered at ClinicalTrials.gov (NCT02568280).

2.2 | Participants

Eligible participants were men and women aged 18 to 64 years, diagnosed with T1D ≥12 months prior to enrolment, treated with multiple daily insulin injections or continuous subcutaneous insulin infusion for ≥12 months (total daily insulin dose <1.2 (I)U/kg/d and total daily bolus insulin dose <0.7 (I)U/kg/d), with a body mass index of 18.5 to 28.0 kg/m^2, a glycated haemoglobin (HbA1c) concentration ≤ 69 mmol/mol (8.5%), fasting C-peptide concentration ≤ 0.3 nmol/L, and with a current, accurate insulin:carbohydrate ratio (defined as 3- to 4-hour PPG increment) <2.8 mmol/L based on all available (and at least 3) self-measured plasma glucose (PG) values before and after breakfast during the last 10 days prior to screening.

2.3 | Procedures

The trial consisted of a screening visit, two dosing visits separated by 3 to 42 days and a follow-up visit. At the two dosing visits, participants received single administration of faster aspart (100 U/mL; Novo Nordisk, Bagsværd, Denmark) or IAsp (NovoRapid® 100 U/mL [Novo Nordisk]) in a randomized sequence. Both trial products were administered subcutaneously into a lifted skin fold of the lower abdominal wall above the inguinal area using a blinded PDS290 pen-injector prefilled pen (Novo Nordisk). The dose was individualized (0.06-0.28 U/kg) based on the participant's customary insulin:carbohydrate ratio and was identical at the two dosing visits for each participant.

At each dosing visit, participants received a standardized dinner at 7:00 PM and were subsequently fasting (except for water intake) until the next day's meal test. A glucose target of 5.5 mmol/L was obtained overnight by variable intravenous (i.v.) infusion of regular human insulin (RHI; Actrapid®, Novo Nordisk) and glucose (10%) starting at 10:00 PM (either insulin or glucose at a given time). The i.v. glucose infusion (if any) was terminated 4 hours prior to the meal test. Water was not allowed within the last 3 hours before the meal test. On the following morning, a 6-hour mixed meal test was conducted and PPG turnover assessed using the triple-tracer method.[10,14] In brief, at 6:00 AM (3 hours before start of the meal test) a primed, continuous, variable i.v. infusion of [6,6-$^2H_2$] glucose was initiated and continued until end of the meal test. At 9:00 AM, the trial product was administered and immediately thereafter, a standardized mixed meal was consumed and completed within 15 minutes. The meal contained 10 kcal/kg body weight (75 g carbohydrate, <40% fat) and consisted of scrambled eggs, meat (lean bacon or steak), butter, reduced fat cheddar cheese, and 75 g glucose labelled with [1-^{13}C] glucose and flavoured with Jell-O (Kraft Foods). At the same time, a continuous, variable i.v. infusion of [6-^{3}H] glucose was initiated. The rate of [6-^{3}H] glucose infusion was adjusted to reflect the expected rate of appearance of [1-^{13}C] glucose originating from the meal, and the rate of [6,6-$^2H_2$] glucose infusion was adjusted to reflect the expected changes in rate of EGP.

Blood samples were drawn frequently up to 6 hours for PG, glucose tracer and free fatty acid (FFA) assessment and up to 8 hours for pharmacokinetic assessment. In case of PG <3.1 mmol/L or in case of hypoglycaemic symptoms irrespective of the PG level, the participant was treated with a dextrose drink (labelled with [1-^{13}C] glucose) to alleviate hypoglycaemia. In case of PG values consistently >19 mmol/L with the presence of hyperglycaemic symptoms, RHI (Actrapid) was to be administered i.v.

Participants did not consume water until 2 hours after dosing (apart from that served with the standardized mixed meal) and did not eat until 6 hours post-dose when they were served meals and snacks. From 6 to 8 hours post-dose, short-acting insulin was limited to RHI.

2.4 | Assessments

Fat-free mass was determined by dual-energy X-ray absorptiometry. Free serum IAsp concentrations (polyethylene glycol precipitated) were determined using a validated IAsp-specific enzyme-linked immunosorbent assay having a lower limit of quantification (LLOQ) of 10 pmol/L.

The PG concentrations were measured using a SuperGL 2 glucose analyser (Dr Müller Gerätebau GmbH, Freital, Germany) using an electrochemical method.

Plasma [6-^{3}H] glucose specific activity was determined using liquid scintillation counting,[14] and plasma enrichment of [1-^{13}C] glucose and [6,6-$^{2}H_2$] glucose was determined by gas chromatography-mass spectrometry (Thermoquest),[15] at the Endocrine Research Unit, Mayo Clinic, Rochester, Minnesota.

Plasma glucagon concentrations were determined in plasma using a validated enzyme-linked immunosorbent assay with an LLOQ of 17.7 pg/mL (Mercodia Glucagon ELISA, Mercodia AB, Uppsala, Sweden).

Safety assessments included adverse events, hypoglycaemic episodes (classified as "severe" according to the American Diabetes Association, ie, requiring third-party assistance,[16] or "confirmed", ie, documented by PG <3.1 mmol/L, with or without symptoms consistent with hypoglycaemia), laboratory safety variables, physical examination, vital signs and ECG.

2.5 | Calculations

Baseline and postprandial rates of glucose turnover were calculated as previously described.[14] In short, the i.v. infusion of [6-^{3}H] glucose was used to trace the rate of appearance of [1-^{13}C] glucose originating from the meal, and the i.v. infusion of [6,6-$^{2}H_2$] glucose was used to trace the rate of EGP. The ratio of plasma concentrations of [6-^{3}H] glucose to [1-^{13}C] glucose was used to calculate the rate of glucose appearance originating from the meal. The rate of EGP and the glucose R_d were then calculated as previously described.[14]

2.6 | Endpoints

Pharmacokinetic endpoints to assess onset of exposure and early exposure were defined and derived as previously described.[7]

The postprandial PG increment during the meal test was assessed by deriving the mean PG excursion from 0 to 1 hour ($\Delta PG_{mean,0\text{-}1h}$; primary endpoint) and from 0 to 2 hours ($\Delta PG_{mean,0\text{-}2h}$) and the PG excursion at 1 and 2 hours (ΔPG_{1h} and ΔPG_{2h}).

Initial EGP suppression during the meal test was assessed by deriving the mean suppression of EGP during the first 30 minutes, 40 minutes and 1 hour (Suppression of $EGP_{0\text{-}x}$). Suppression of $EGP_{0\text{-}x}$ was calculated as baseline-adjusted area over the EGP curve from time 0 to *x* divided by the time period *x* (to obtain mean baseline-adjusted EGP) and then divided by baseline EGP and expressed in percent. EGP suppression from baseline until a discrete time point (suppression of EGP_x) was calculated as baseline EGP minus EGP at time *x* divided by baseline EGP and expressed in percent. Maximum EGP suppression was calculated as baseline EGP minus the minimum EGP divided by baseline EGP and expressed in percent.

The initial increase in glucose R_d during the meal test was assessed by deriving the area under the baseline-adjusted glucose R_d profile during the first hour ($\Delta AUC_{Rd,0\text{-}1h}$). The baseline glucose R_d was set to baseline EGP, since glucose R_d equals EGP in the fasting state.

The initial suppression of serum FFA concentration during the meal test was assessed by deriving the area over the baseline-adjusted serum FFA concentration–time curve during the first hour ($\Delta AOC_{FFA,0\text{-}1h}$).

The mean plasma glucagon concentration was presented graphically per treatment. The individual plasma glucagon concentrations were below the LLOQ of 17.7 pg/mL for a number of measurement time points. These values were set to zero. Because of the number of individual plasma glucagon concentrations below the LLOQ, it was not possible to calculate endpoints for glucagon.

2.7 | Statistical analysis

Assuming a true treatment difference of 0.85 mmol/L for the primary endpoint, $\Delta PG_{mean,0\text{-}1h}$, and a within-participant standard deviation of 1 mmol/L (from a previous trial with faster aspart and IAsp[17]), 31 completing participants were required to show a statistically significant treatment difference with 90% power when using a two-sided test and a significance level of 5%. In order to take non-completers into account, 38 participants were planned to be randomized in the trial.

Endpoints were compared between treatments in a linear model, with treatment, period and participant as fixed effects. For analysis of $\Delta PG_{mean,0\text{-}1h}$, $\Delta PG_{mean,0\text{-}2h}$, ΔPG_{1h}, ΔPG_{2h} and $\Delta AOC_{FFA,0\text{-}1h}$, the pre-dose value was included as covariate. Early and overall exposure endpoints were log-transformed prior to analysis. For endpoints analysed on the original scale, treatment ratios and 95% confidence intervals (CIs) were calculated by Fieller's method.[18]

3 | RESULTS

3.1 | Participant disposition and baseline characteristics

A total of 76 individuals were screened, 42 were randomized, and 41 were exposed to the trial products and completed the trial. One participant was withdrawn before exposure because of a lack of PG stabilization before trial product administration. The safety analysis set included the 41 exposed participants. The full analysis set (used for pharmacokinetic analyses) included 40 participants (one participant who received a different dose at the two dosing visits was excluded). Pharmacodynamic analyses included 38 participants (2 participants ingesting a different meal size at the two dosing visits were excluded). Participant disposition is presented in Figure S1. Participant characteristics are shown in Table 1.

3.2 | Pharmacokinetics

The mean pharmacokinetic profile was shifted to the left for faster aspart vs IAsp (Figure 1A). Accordingly, faster aspart provided earlier

TABLE 1 Participant characteristics

	Participants with T1D *N* = 40
Age, years	42.0 (12.1)
Sex	
Women, *n* (%)	21 (52.5)
Men, *n* (%)	19 (47.5)
Race	
White, *n* (%)	39 (97.5)
Asian, *n* (%)	1 (2.5)
Body weight, kg	72.4 (10.8)
Fat-free mass, kg	52.1 (10.1)
BMI, kg/m^2	24.1 (2.2)
Duration of diabetes, years	19.5 (11.6)
HbA1c	
mmol/mol	56 (8)
%	7.3 (0.7)

Abbreviations: BMI, body mass index; HbA1c, glycated haemoglobin; *n*, number of subjects; T1D, type 1 diabetes. Data are mean (SD) unless otherwise stated.

onset of exposure as well as greater initial exposure after subcutaneous administration compared with IAsp. Thus, shorter $t_{Early\ 50\%\ Cmax}$ (by 9 minutes; $P < .001$) and earlier t_{max} (by 19 minutes; $P < .001$) were seen with faster aspart vs IAsp (Table 2). Early exposure within the first 2 hours after administration was statistically significantly greater for faster aspart vs IAsp (Figure 2). During the first 15, 30 and 60 minutes after administration, respectively, ~3.7-fold greater, ~2-fold greater and 32% greater insulin exposure was seen with faster aspart than with IAsp ($P < .001$).

Total exposure ($AUC_{IAsp,0\text{-}t}$) and maximum concentration (C_{max}) were similar for faster aspart and IAsp. LS means (between-participant coefficient of variation in %) for $AUC_{IAsp,0\text{-}t}$ were 370 pmol·h/L (1%) and 378 pmol·h/L (1%) for faster aspart and IAsp, respectively (estimated ratio faster aspart/IAsp 0.98 [95% CI 0.95;1.01]; $P = .141$). LS means for C_{max} were 171 pmol/L (3%) and 162 pmol/L (3%) for faster aspart and IAsp, respectively (1.06 [95% CI 0.97; 1.15]; $P = .190$).

3.3 | Pharmacodynamics

Glucose intervention with a dextrose drink (labelled with [1-^{13}C] glucose) to alleviate hypoglycaemia during the 6-hour meal test occurred in 5 participants with faster aspart and 8 participants with IAsp. No glucose intervention occurred within the first 2 hours of the meal test. Thus, glucose intervention during the meal test did not influence the presented pharmacodynamic endpoints. No interventions were needed to alleviate hyperglycaemia.

Over the first 2 hours of the meal test, an apparently smaller increase in PG was seen for faster aspart vs IAsp, with the greatest difference observed during the first hour (Figure 1B). $\Delta PG_{mean,0\text{-}1h}$ (primary endpoint) and ΔPG_{1h} showed trends towards a greater reduction with faster aspart than with IAsp (Table 2). $\Delta PG_{mean,0\text{-}2h}$ and ΔPG_{2h} did not differ statistically significantly between faster aspart and IAsp.

Mean profiles of tracer-to-tracee ratios for [6-^{3}H] glucose/[1-^{13}C] glucose (used to calculate the rate of meal glucose appearance) and [6,6-^{2}H$_2$] glucose/endogenous glucose (used to calculate EGP) are shown in Figure S2.

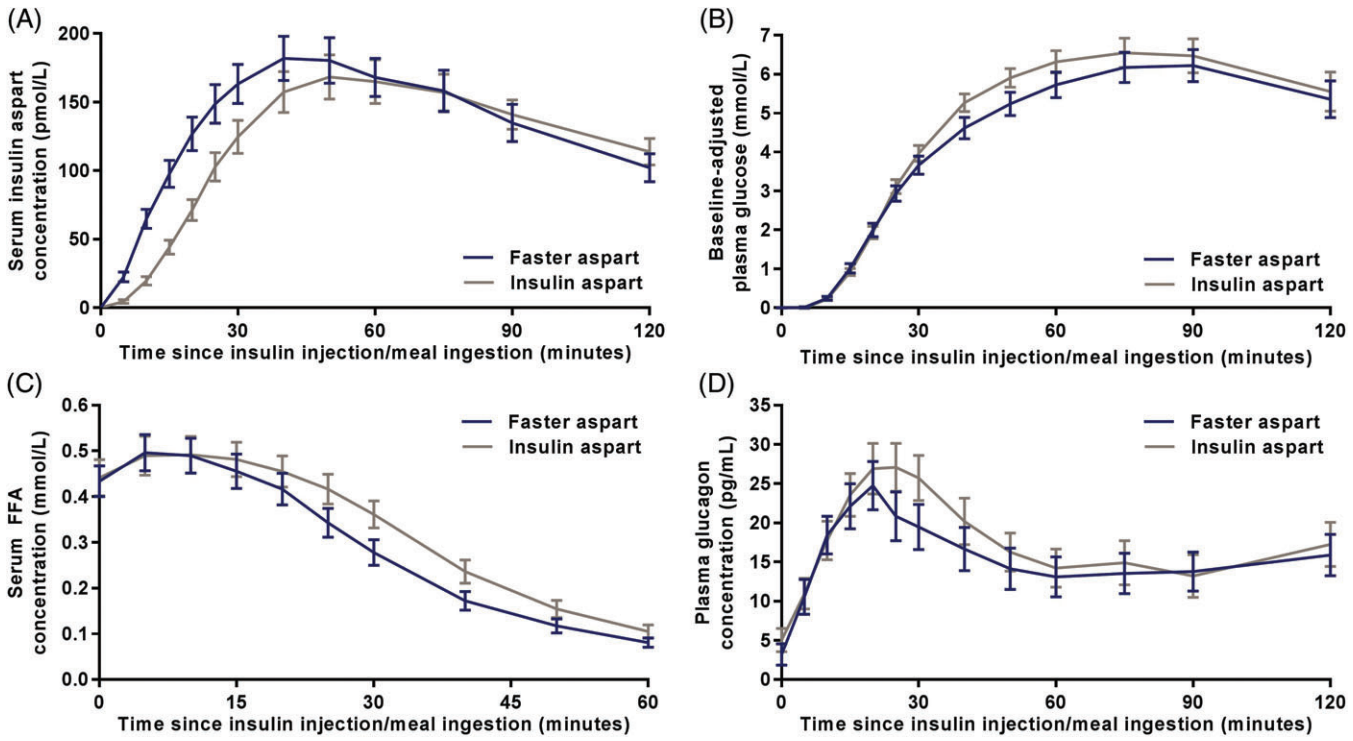

FIGURE 1 Mean serum insulin concentration (A), mean baseline-adjusted plasma glucose concentration (B), mean serum free fatty acid (FFA) concentration (C), and mean plasma glucagon concentration (D), for fast-acting insulin aspart (faster aspart) and insulin aspart during a meal test after individualized subcutaneous dosing (0.06-0.28 U/kg) in participants with type 1 diabetes. Error bars show SEM

TABLE 2 Onset of exposure and postprandial glucose increment for fast-acting insulin aspart vs insulin aspart during a meal test after individualized subcutaneous dosing (0.06-0.28 U/kg) in participants with type 1 diabetes

Onset of exposure	Faster aspart[a], min	IAsp[a], min	Treatment ratio[b] (95% CI)	Treatment difference[c] (95% CI), min	*P*[d]
$t_{Early\ 50\%\ Cmax}$	16.9 ± 0.8	25.5 ± 0.8	0.66 (0.58;0.75)	−8.7 (−11.1;−6.2)	<.001
t_{max}	50.5 ± 3.4	69.3 ± 3.4	0.73 (0.61;0.86)	−18.8 (−28.5;−9.0)	<.001
PPG increment	**Faster aspart[a], mmol/L**	**IAsp[a], mmol/L**	**Treatment ratio[b] (95% CI)**	**Treatment difference[c] (95% CI), mmol/L**	***P*[d]**
$\Delta PG_{mean,0\text{-}1h}$[e]	3.09 ± 0.12	3.40 ± 0.12	0.91 (0.81;1.02)	−0.31 (−0.66;0.05)	.089
ΔPG_{1h}	5.73 ± 0.21	6.32 ± 0.21	0.91 (0.82;1.00)	−0.59 (−1.19;0.01)	.055
$\Delta PG_{mean,0\text{-}2h}$	4.51 ± 0.18	4.82 ± 0.18	0.94 (0.83;1.05)	−0.31 (−0.84;0.22)	.245
ΔPG_{2h}	5.36 ± 0.30	5.55 ± 0.30	0.97 (0.82;1.13)	−0.19 (−1.05;0.66)	.646

Abbreviations: $\Delta PG_{mean,0\text{-}xh}$, mean postprandial plasma glucose increment from 0 to *x* hours; ΔPG_{xh}, postprandial plasma glucose increment at *x* hours; PPG, postprandial glucose; $t_{Early\ 50\%\ Cmax}$, time to 50% of maximum insulin concentration in the early part of the pharmacokinetic profile; t_{max}, time to maximum insulin concentration.

[a] Data are least squares means ± SEM.

[b] Faster aspart/IAsp (calculated using Fieller's method).

[c] Faster aspart – IAsp.

[d] For the comparison of faster aspart vs IAsp.

[e] Primary endpoint.

The rate of glucose appearance originating from the meal was similar for faster aspart and IAsp during the first hour of the meal test (Figure S3), thereby simplifying the interpretation of the other pharmacodynamic variables.

The baseline-adjusted rate of EGP over the first hour of the meal test is shown in Figure S4, which indicates a greater suppression of EGP with faster aspart vs IAsp until 30 to 40 minutes after meal ingestion. Statistical analysis showed that the suppression of EGP was twice as large during the first 30 minutes (*P* = .017), 34% greater during the first 40 minutes (*P* = .019) and 12% greater during the first hour (*P* = .040) of the meal test with faster aspart vs IAsp (Figure 3A).

The EGP suppression at 30 minutes, 40 minutes and 1 hour relative to baseline is presented in Figure 3B, showing 33% greater EGP suppression at 30 minutes with faster aspart vs IAsp (*P* = .049), while no statistically significant treatment difference was seen at 40 minutes (*P* = .247) and 1 hour (*P* = .219). Maximum suppression of EGP did not differ statistically significantly between faster aspart and IAsp, with LS means±standard error of 68.4 ± 0.93% and 68.2 ± 0.93%, respectively, and an estimated treatment ratio of 1.00 (95% CI 0.96;1.04; *P* = .906).

Based on the mean profiles of glucose R_d (Figure S5) and serum FFA concentration (Figure 1C) during the first hour of the meal test, a greater increase in glucose R_d was observed until 40 to 50 minutes after meal ingestion, and a greater suppression of serum FFA concentration was observed until ~1 hour after meal ingestion, with faster aspart compared with IAsp. $\Delta AUC_{Rd,0\text{-}1h}$ was 23% greater (*P* = .012) and $\Delta AOC_{FFA,0\text{-}1h}$ was 36% greater (*P* = .042) for faster aspart vs IAsp (Figure 3C). No major treatment differences were seen in glucose R_d and serum FFA concentration beyond 1 hour (data not shown).

Plasma glucagon concentration over the first 2 hours of the meal test is presented in Figure 1D, indicating a slightly lower level of circulating glucagon from ~15 minutes until 1 hour after meal ingestion for faster aspart vs IAsp.

3.4 | Safety

Faster aspart and IAsp were well tolerated, and no safety issues were identified. A total of 6 adverse events (5 after faster aspart and 1 after IAsp) were reported in 5 participants, which were mainly of mild intensity and were assessed to be unrelated to the trial product. All participants recovered from the events. There were no serious adverse events. A total of 6 confirmed hypoglycaemic episodes (1 after faster aspart and 5 after IAsp) were reported in 6 participants (none were severe and none occurred within the first 2 hours

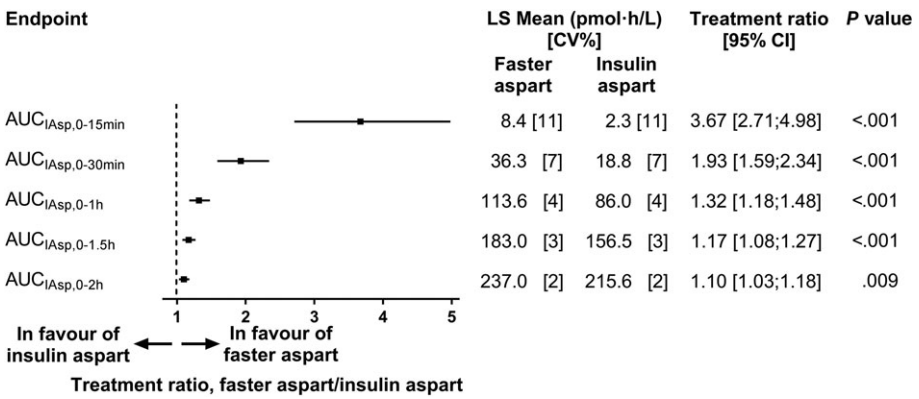

FIGURE 2 Early exposure for fast-acting insulin aspart (faster aspart) vs insulin aspart during a meal test after individualized subcutaneous dosing (0.06-0.28 U/kg) in participants with type 1 diabetes. AUC, area under the curve; CV%, between-participant coefficient of variation in %; IAsp, insulin aspart; LS Mean, least squares mean; *P* value, treatment comparison of faster aspart vs IAsp; treatment ratio, faster aspart/IAsp

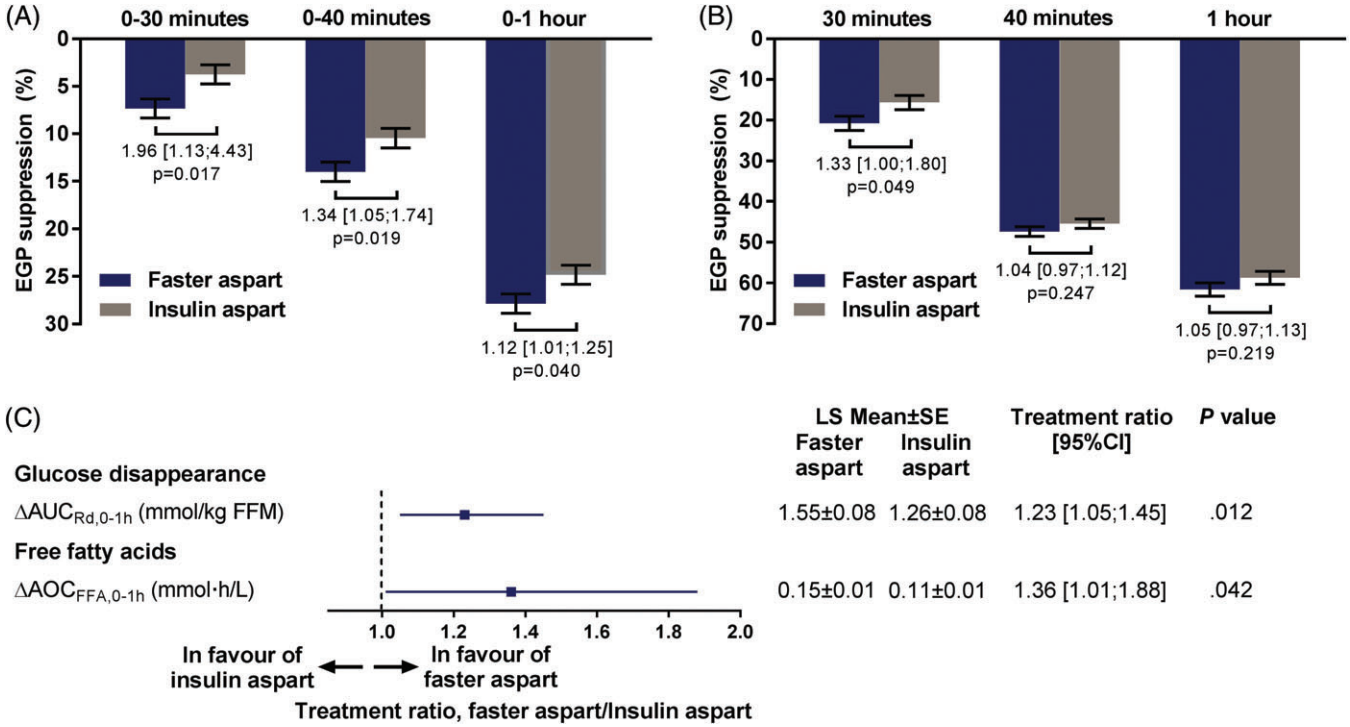

FIGURE 3 Suppression of endogenous glucose production (EGP; A and B), increase in glucose disappearance and decrease in free fatty acids (FFA; C) for fast-acting insulin aspart (faster aspart) versus insulin aspart during a meal test after individualized subcutaneous dosing (0.06-0.28 U/kg) in participants with type 1 diabetes. A, Mean suppression of EGP over the indicated time periods. B, EGP suppression from baseline until the discrete time points as indicated. In A and B, bars are LS means ± SE and treatment comparisons show treatment ratios of faster aspart/insulin aspart [95% CI] and *P* value. AOC, area over the curve; AUC, area under the curve; FFM, fat-free mass; LS Mean, least squares mean; *P* value, treatment comparison of faster aspart vs insulin aspart; R_d, rate of glucose disappearance; treatment ratio, faster aspart/insulin aspart

post-dose). There were no clinically significant observations in safety laboratory variables, vital signs, physical examination or ECG results.

4 | DISCUSSION

To the best of our knowledge, the present trial is the first to compare PPG fluxes between two exogenous short-acting insulin products with different pharmacological profiles. Since the pharmacological profiles of faster aspart and IAsp are known to differ mainly within the first 1 to 2 hours after administration,[6] the present trial focused on that time period. The main finding was that within the first hour of meal ingestion, faster aspart administration led not only to higher glucose R_d but also to greater suppression of EGP compared with IAsp. These effects of faster aspart on glucose turnover collectively resulted in a trend towards a reduced PPG increment with faster aspart vs IAsp. Thus, the present trial provides the mechanism(s), regarding glucose fluxes, behind the reduction in PPG increment with faster aspart compared with IAsp shown in recent clinical trials in individuals with T1D and T2D.[8,9]

The relative contributions from treatment differences in postprandial glucose R_d and EGP suppression to the smaller ΔPG_{1h} with faster aspart were not directly estimated in the present trial; however, it can be inferred by comparison of the areas between the curves in Figures S4 and S5 that the increased stimulation of glucose R_d with faster aspart vs IAsp was the most important contributor to the treatment difference in ΔPG_{1h}. Nevertheless, the finding of greater early EGP suppression with faster aspart vs IAsp is highly interesting. Exogenous insulin products administered subcutaneously are likely to shift the normal hepatic to peripheral insulin gradient, thereby causing relative peripheral hyperinsulinaemia and under-insulinization of the liver.[19] It is therefore reassuring that part of the improved PPG control with faster aspart is attributable to effects exerted on the liver which may, at least partly, be attributable to greater early suppression of postprandial circulating FFA concentration, another finding in the present trial (Figure 1C). While comparison of glucose turnover results between studies are fraught with considerable limitations, it is intriguing that the earlier exposure to insulin with faster aspart still does not fully restore insulin stimulation of glucose R_d nor insulin suppression of EGP to the rates observed in healthy individuals without diabetes.[13]

The maximum postprandial EGP suppression of 68% is in line with previous findings.[20] The regulation of postprandial EGP suppression is complex and depends mainly on circulating insulin, glucose and glucagon concentrations.[21] This may explain why some variability in EGP between two consecutive mornings has been found in individuals with T1D.[22] In the present trial, however, the greater postprandial EGP suppression with faster aspart vs IAsp was sufficiently robust to show statistically significant differences at all three time periods assessed (Figure 3A).

Although the accelerated pharmacokinetic profile of faster aspart vs IAsp was presumably the primary reason for the greater early postprandial EGP suppression with faster aspart, other indirect factors may also have played a role. In patients with T1D, the lack of

postprandial insulin secretion from the pancreatic β cells leads to a paradoxical increase in glucagon concentration in response to a meal, contrary to what is seen in healthy individuals.[23] It has been shown both in T1D and T2D that lack of postprandial suppression of glucagon contributes to hyperglycaemia via elevated hepatic glucose release.[24,25] In the present trial, the postprandial increase in glucagon was slightly less for faster aspart than for IAsp (Figure 1D), which may have contributed to the greater EGP suppression with faster aspart; however, given that individual plasma glucagon concentrations were below the LLOQ at several time points, as mentioned earlier, we cannot be certain of the contribution of lower plasma glucagon concentrations on EGP with faster aspart. Furthermore, the lower postprandial circulating FFA concentration with faster aspart vs IAsp may have improved the ability of insulin to suppress EGP and may also have reduced the availability of FFA to the liver, thereby limiting FFA oxidation and thus the rate of gluconeogenesis.[21]

The fraction of glucose derived from the meal, taken up by the liver during the first pass from the gut via the portal vein into the systemic circulation, constitutes another component of postprandial hepatic glucose turnover. This component was not measured in the present trial, as this would have required hepatic vein catheterization; however, because the rate of systemic appearance of glucose originating from the meal was similar for faster aspart and IAsp during the first hour of the meal test (Figure S3) and beyond (data not shown), the first-pass hepatic glucose uptake was also most likely similar for faster aspart and IAsp, and therefore apparently not influenced by the faster onset and greater early exposure seen with faster aspart.

The most important strength of the present trial was the use of the triple-tracer technique, which limited non-steady-state errors by minimizing changes in the tracer-to-tracee ratios used to measure meal glucose rate of appearance and EGP.[10,14] As shown in Figure S2, the ratio of [6,6-$^{2}H_2$] glucose to endogenous glucose was constant and unchanging throughout the duration of the experiment during both study visits, thereby implying that the rates of calculated EGP are robust. However, as has been observed in prior triple-tracer meal studies, the ratio of [6-^{3}H] glucose to [1-^{13}C] glucose, used to calculate meal glucose rate of appearance, varied for the first 30 minutes after meal ingestion, then became relatively smooth for the next 2 hours before gradually rising for the rest of the experiment congruently during both study visits. The imprecision of measurement of meal glucose rate of appearance for the initial postprandial period, therefore, appears to be similarly affected during both study visits. Still, the non-steady-state situation early after meal ingestion and insulin administration, together with a minor transient increase in EGP, implied that it was not valid to derive endpoints related solely to very early EGP suppression (up to 15 minutes) and glucose R_d (up to 30 minutes). Another strength was the use of an individualized prandial insulin dose, thereby reflecting clinical practice. While the current highly standardized experimental setup ensured robust conclusions on the mechanisms behind the trend towards a lower PPG increment with faster aspart, it could also be a limitation. For example, the standard mixed meal ingested by all participants puts certain limitations on the clinical applicability of the results. Along these lines, the present findings should also be interpreted in light of the fact that the reduction in PPG increment with faster aspart vs IAsp in the present trial (−0.59 mmol/L) was less than observed in a recent phase III trial in participants with T1D (−1.18 mmol/L).[8] The present study was conducted in participants with T1D. Using the triple-tracer technique in participants with T2D, we have previously demonstrated significant postprandial hepatic and peripheral insulin resistance in these individuals.[13] Future studies are therefore necessary in insulin-requiring people with T2D to determine the extent to which, if at all, faster aspart alters insulin suppression of EGP and stimulation of glucose R_d in that population.

In conclusion, in line with previous findings, the present trial showed that faster aspart provides earlier onset of exposure and greater early exposure compared with IAsp, which in the present study, led to a trend towards improved 1-hour PPG control with faster aspart. This trend was attributable to greater suppression of EGP as well as higher glucose R_d with faster aspart vs IAsp.

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Declaration of interests

A.B. has received research support from AstraZeneca, Dexcom and Tandem Diabetes Care. T.R.P. has received research support from AstraZeneca and Novo Nordisk, has served on advisory panels for AstraZeneca, Bristol-Myers Squibb, Eli Lilly, Novo Nordisk and Roche Diabetes Care and is an employee of CBmed - Center for Biomarker Research in Medicine (a public owned research company). R.B. has received research support from AstraZeneca and Novo Nordisk. A.K.H., L.E. and H.H. are employees and shareholders of Novo Nordisk. S. S.-F. declares no conflicts of interest.

Author contributions

A.B., T.R.P., A.K.H. and R.B. contributed to study design, conduct/data collection, analysis and writing the manuscript. S.S.-F. contributed to conduct/data collection, analysis and writing the manuscript. L.E. contributed to analysis and writing the manuscript. H.H. contributed to study design, analysis and writing the manuscript.

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REFERENCES

1. Monnier L, Lapinski H, Colette C. Contributions of fasting and postprandial plasma glucose increments to the overall diurnal hyperglycemia of type 2 diabetic patients: variations with increasing levels of HbA_{1c}. *Diabetes Care*. 2003;26:881-885.
2. Ketema EB, Kibret KT. Correlation of fasting and postprandial plasma glucose with HbA1c in assessing glycemic control; systematic review and meta-analysis. *Arch Public Health*. 2015;73:43.
3. Woerle HJ, Neumann C, Zschau S, et al. Impact of fasting and postprandial glycemia on overall glycemic control in type 2 diabetes. Importance of postprandial glycemia to achieve target HbA1c levels. *Diabetes Res Clin Pract*. 2007;77:280-285.

4. Heise T, Hövelmann U, Brøndsted L, Adrian CL, Nosek L, Haahr H. Faster-acting insulin aspart: earlier onset of appearance and greater early pharmacokinetic and pharmacodynamic effects than insulin aspart. *Diabetes Obes Metab*. 2015;17:682-688.
5. Buckley ST, Kildegaard J, Høiberg-Nielsen R, et al. Mechanistic analysis into the mode(s) of action of niacinamide in faster-acting insulin aspart (Abstract). *Diabetes Technol Ther*. 2016;18(suppl 1):A116-A117.
6. Heise T, Pieber TR, Danne T, Erichsen L, Haahr H. A pooled analysis of clinical pharmacology trials investigating the pharmacokinetic and pharmacodynamic characteristics of fast-acting insulin aspart in adults with type 1 diabetes. *Clin Pharmacokinet*. 2017;56:551-559.
7. Heise T, Stender-Petersen K, Hövelmann U, et al. Pharmacokinetic and pharmacodynamic properties of faster-acting insulin aspart versus insulin aspart across a clinically relevant dose range in subjects with type 1 diabetes mellitus. *Clin Pharmacokinet*. 2017;56:649-660.
8. Russell-Jones D, Bode B, de Block C, et al. Fast-acting insulin aspart improves glycemic control in basal-bolus treatment for type 1 diabetes: results of a 26-week multicenter, active-controlled, treat-to-target, randomized, parallel-group trial (Onset 1). *Diabetes Care*. 2017;40:943-950.
9. Bowering K, Case C, Harvey J, et al. Faster aspart versus insulin aspart as part of a basal-bolus regimen in inadequately controlled type 2 diabetes: the onset 2 trial. *Diabetes Care*. 2017;40:951-957.
10. Rizza RA, Toffolo G, Cobelli C. Accurate measurement of postprandial glucose turnover: why is it difficult and how can it be done (relatively) simply? *Diabetes*. 2016;65:1133-1145.
11. Saad A, Dalla Man C, Nandy DK, et al. Diurnal pattern to insulin secretion and insulin action in healthy individuals. *Diabetes*. 2012;61:2691-2700.
12. Rizza RA. Pathogenesis of fasting and postprandial hyperglycemia in type 2 diabetes: implications for therapy. *Diabetes*. 2010;59:2697-2707.
13. Basu A, Dalla Man C, Basu R, Toffolo G, Cobelli C, Rizza RA. Effects of type 2 diabetes on insulin secretion, insulin action, glucose effectiveness, and postprandial glucose metabolism. *Diabetes Care*. 2009;32:866-872.
14. Basu R, Di Camillo B, Toffolo G, et al. Use of a novel triple-tracer approach to assess postprandial glucose metabolism. *Am J Physiol*. 2003;284:E55-E69.
15. Beylot M, Previs SF, David F, Brunengraber H. Determination of the 13C-labelling pattern of glucose by gas chromatography-mass spectrometry. *Anal Biochem*. 1993;212:526-531.
16. American Diabetes Association. Defining and reporting hypoglycaemia in diabetes: a report from the American Diabetes Association Workgroup on Hypoglycaemia. *Diabetes Care*. 2005;28:1245-1249.
17. Heise T, Haahr H, Jensen L, Erichsen L, Hompesch M. Faster-acting insulin aspart improves postprandial glycemia vs insulin aspart in patients with type 1 diabetes mellitus (T1DM) (Abstract). *Diabetes*. 2014;63(suppl 1):A34.
18. Fieller EC. Some problems in interval estimation. *J R Stat Soc Series B Stat Methodol*. 1954;16:175-185.
19. Herring R, Jones RH, Russell-Jones DL. Hepatoselectivity and the evolution of insulin. *Diabetes Obes Metab*. 2014;16:1-8.
20. Hinshaw L, Schiavon M, Dadlani V, et al. Effect of pramlintide on postprandial glucose fluxes in type 1 diabetes. *J Clin Endocrinol Metab*. 2016;101:1954-1962.
21. Consoli A. Role of liver in pathophysiology of NIDDM. *Diabetes Care*. 1992;15:430-441.
22. Mallad A, Hinshaw L, Dalla Man C, et al. Nocturnal glucose metabolism in type 1 diabetes: a study comparing single versus dual tracer approaches. *Diabetes Technol Ther*. 2015;17:587-595.
23. Cryer PE. Minireview: glucagon in the pathogenesis of hypoglycemia and hyperglycemia in diabetes. *Endocrinology*. 2012;153:1039-1048.
24. Dinneen S, Alzaid A, Turk D, Rizza R. Failure of glucagon suppression contributes to postprandial hyperglycaemia in IDDM. *Diabetologia*. 1995;38:337-343.
25. Shah P, Vella A, Basu A, Basu R, Schwenk WF, Rizza RA. Lack of suppression of glucagon contributes to postprandial hyperglycemia in subjects with type 2 diabetes mellitus. *J Clin Endocrinol Metab*. 2000;85:4053-4059.

SUPPORTING INFORMATION

Additional Supporting Information may be found online in the supporting information tab for this article.

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