available at www.sciencedirect.com

ScienceDirect

journal homepage: www.elsevier.com/locate/diabres

A comparison of preprandial insulin glulisine versus insulin lispro in people with Type 2 diabetes over a 12-h period☆

Stephen Luzio *, *Rajesh Peter*, *Gareth J. Dunseath*, *Laila Mustafa*, *David R. Owens*

Diabetes Research Unit, Llandough Hospital, South Glamorgan, United Kingdom

ARTICLE INFO

Article history:
Received 5 June 2007
Accepted 22 November 2007
Published on line 27 December 2007

Keywords:
Glulisine
Lispro
Pharmacokinetics
Pharmacodynamics
Type 2 diabetes

ABSTRACT

A comparison of the plasma glucose and insulin day profiles between two prandial rapid-acting insulin analogues, insulin glulisine (glulisine) and insulin lispro (lispro), in 18 obese subjects with Type 2 diabetes.

Subjects (body mass index: males, 36.7 [33.2–43.8] kg/m^2; females, 40.0 [35.7–46.5] kg/m^2) received subcutaneous glulisine or lispro (0.15 U/kg) at 4-h intervals immediately (within 2 min) before three standard test meals during each of two 12-h, randomised, open-label, crossover studies (7 ± 2-day interval between each).

Overall, preprandial-subtracted glucose concentrations (area under the curve) were similar on the glulisine and lispro study days. However, the mean of the three maximal preprandial subtracted plasma glucose concentrations (ΔGLU_{max}) were lower with glulisine versus lispro (12%; $p < 0.01$). Mean concentrations of insulin analogue were significantly higher post-meal with glulisine ($p < 0.01$ for all). *Post hoc* analysis showed a significantly faster absorption rate for glulisine versus lispro in the first 30 min post-meal (estimated difference 0.48 μU/min; $p < 0.0001$). Only two cases of hypoglycaemia were reported; both from one subject during the lispro day.

When glulisine is injected immediately before a meal in obese patients with Type 2 diabetes, glulisine achieves significantly lower glucose excursions over lispro. Significantly faster absorption with higher and sustained post-meal levels of insulin analogue was achieved at every meal with glulisine versus lispro.

1. Introduction

The primary goal of insulin therapy in Type 2 diabetes mellitus (T2DM) is to achieve tight glycaemic control by supplementing the insulin deficit in a manner that is as close to the normal insulin secretion pattern as possible. The key features of a normal insulin profile involve a sustained and relatively constant basal level of insulin secretion, along with a meal-stimulated peak (30–60 min) of insulin secretion that slowly decays over the subsequent 2–3 h. Basal-bolus insulin therapy, involving the use of a combination of rapid- and long-acting insulin analogue preparations replicates this pattern and provides a physiological form of insulin replacement therapy [3]. Although regular human insulin (RHI) has traditionally been used as bolus (prandial) insulin, its pharmacological profile does not resemble the profile of endogenous insulin

☆ This study was funded and sponsored by sanofi-aventis. Data from this manuscript has previously been presented at the European Association for the Study of Diabetes 2005 meeting, 12–15 September, Athens, Greece [1] and the Diabetes UK Annual Professional Conference 2006, 29–31 March, Birmingham, UK [2].

* *Corresponding author at*: Diabetes Research Unit, Llandough Hospital, Penlan Road, Penarth CF64 2XX, United Kingdom. Tel.: +44 2920716921; fax: +44 2920350147.
E-mail address: luzio@Cardiff.ac.uk (S. Luzio).

0168-8227/$ – see front matter
doi:10.1016/j.diabres.2007.11.013

release [4]. It has an onset of action of 30 min and a duration of action of 6–8 h—resulting in a recommendation that it must be given at least 30 min prior to a meal [5,6].

Insulin glulisine (glulisine) is a new rapid-acting insulin analogue developed to more closely resemble physiological insulin release after meals and, therefore, improve prandial glycaemic control [7]. Glulisine differs in structure from RHI by the replacement of asparagine with lysine at position 3 and of lysine with glutamic acid at position 29 on the B chain of the human insulin molecule [8]. It has previously been demonstrated that glulisine provides better glycaemic (HbA_{1c}) control versus RHI in patients with Type 1 diabetes mellitus (T1DM) when administered 0–15 min pre-meal and equivalent control when given immediately after the meal [7]. Furthermore, when compared with insulin lispro (lispro), glulisine shows equivalent glycaemic control in patients with T1DM [9]. Similarly, glulisine displays a more rapid onset and a shorter duration of action when compared with RHI in patients with T2DM [10], which is associated with improved HbA_{1c} and lower post-breakfast and post-dinner blood glucose levels versus RHI [11].

Obesity is frequently associated with T2DM [12,13] and it is thus important that insulin analogues maintain their pharmacokinetic (PK) and pharmacodynamic (PD) characteristics regardless of body fat, skin thickness or body mass index (BMI). However, increasing the subcutaneous (sc) fat layer at the injection site may delay the rate of insulin absorption [14]. A study of obese subjects without diabetes showed that increasing skin thickness (sc fat layer) and BMI had a detrimental effect on the PK profiles of both lispro and RHI, whereas the time-action profile of glulisine was less affected [15]. There was a positive correlation between skin thickness or BMI and PD parameters for lispro and RHI. In contrast, the time-action profile of glulisine did not demonstrate any significant correlation with either anthropometric measure and may provide advantages to patients with T2DM. The present exploratory study in obese patients with T2DM compared the PK and PD profiles of glulisine with lispro when administered sc before three standard meals during a 12-h period.

2. Materials and methods

2.1. Study population

Male and female obese subjects (BMI $> 30\ kg/m^2$) aged 18–75 years with T2DM and treated with oral hypoglycaemic agents (OHAs) for at least 6 months were included in the study. Eighteen out of 22 subjects screened were randomised to treatment; nine subjects received lispro in trial period 1, followed by glulisine in trial period 2; and nine received glulisine in trial period 1, followed by lispro in trial period 2. One female subject was replaced by a male subject during the study due to an adverse event, which was considered to be unrelated to the study medication. In total, 18 subjects completed the crossover study according to the protocol and were included in the statistical analyses. All 19 subjects who received the study treatment were included in the safety analyses.

The choice of sample size was based on previous experience and, owing to the exploratory nature of the study, formal sample size calculation was not performed. The study was conducted between November and December 2004 in a single centre, in accordance with the Principles of Good Clinical Practice, the UK Medicines for Human Use (Clinical Trials) Act 2004 and the Declaration of Helsinki. Approval by an independent ethics committee was obtained for the study centre and all patients provided written informed consent prior to study entry.

2.2. Study design

This was a randomised, open-label, two-arm, crossover study with four trial periods: screening (trial period 0), two 12-h treatment visits (trial periods 1 and 2) and a follow-up visit (trial period 3). During trial period 1, subjects were randomised via a sponsor-generated randomisation schedule (sent with numbered containers containing the study medication) to receive either glulisine or lispro. The physician checked that the correct medication was given to each subject according to the schedule. After a washout period of 7 ± 2 days, subjects received the alternative insulin analogue treatment during trial period 2.

2.3. Study protocol

During the whole study period, with the exception of the 12-h study days, subjects were treated with their usual OHAs, the dose of which remained fixed throughout the study period. The night before the 12-h study day, all OHA treatment, with the exception of metformin, was stopped. Subjects were fasted (but allowed to drink water) for 10 h prior to each 12-h study day, before admission to the Diabetes Investigation Unit. On arrival at the clinic, a cannula was inserted into the forearm to enable blood sampling, with a saline-infusion tap attached to maintain patency of the vein. Blood samples were taken at pre-defined intervals before administration of the study medication, and for 4 h after each injection for the measurement of plasma glucose, C-peptide and serum insulin.

During the 12-h study day, subjects received three doses of glulisine or lispro (0.15 U/kg) given sc into the anterior abdominal wall at 4-h intervals immediately prior (within 2 min) to three 500 kcal (58% carbohydrate, 20% protein and 22% fat) standard test meals (breakfast, lunch and dinner) given at approximately 08:00, 12:00 and 16:00 h. Subjects were only allowed to drink water between each standardised meal. The injection site was assessed for any adverse reactions after each injection of the study medication.

2.4. Analytical methods

Plasma glucose concentrations were determined using a hexokinase assay (BioStat Ltd., Stockport, UK). Concentrations of each insulin analogue were assayed by radioimmunoassay methods specific to glulisine and lispro (Linco Research Inc., Missouri, USA). Both assays went through a validation procedure to ensure approximately 100% cross-reactivity with the respective insulin analogue and little or no cross-reactivity with human insulin or proinsulin. These validation procedures ensured broad comparability of the assays; however, since different antibodies and standards were used in each specific assay, it was not possible to ensure that the assays were exactly comparable.

Concentrations of C-peptide were determined using a C-peptide immunochemiluminometric assay (MLT Research Ltd., Cardiff, UK). In addition, injection site skin thickness was measured using standard ultrasound techniques.

2.5. *Statistical analyses*

2.5.1. *Pharmacodynamics*

All areas under the curve (AUCs), maximum plasma glucose concentrations (GLU_{max}), minimum plasma glucose concentrations (GLU_{min}) and maximum preprandial-subtracted plasma glucose concentrations (ΔGLU_{max}) were analysed using mixed models (between and within subject factors) of analysis of variance (ANOVA). Results were expressed as the ratio of geometric means for each insulin analogue parameter accompanied by 90% confidence intervals (CIs), derived using Fieller's theorem. The time to GLU_{max} ($GLU\text{-}T_{max}$) was analysed using ANOVA; 90% non-parametric CIs for the respective median differences in treatment were calculated.

2.5.2. *Pharmacokinetics*

Data were assessed to see if they were normally distributed. For data that were not normally distributed, ANOVA was performed on ln-transformed data for each timepoint (AUC data) and on the maximum insulin analogue concentration ($INS\text{-}C_{max}$). Additional terms for time, and the interaction between time and treatment were also fitted. Random effects models were used for all analyses.

The time to $INS\text{-}C_{max}$ ($INS\text{-}T_{max}$) was measured using the same methods as for $INS\text{-}C_{max}$; however, these data were not ln-transformed, so the estimated treatment difference was in the form of a difference between arithmetic means rather than a ratio.

The absorption rate of insulin analogue for the first 30 min after each meal (at 0, 10, 20 and 30 min) was estimated using a regression line fitted to the insulin analogue concentrations for each subject during each period.

2.5.3. *Other analyses*

The relationships between the PK and PD variables and skin thickness were investigated using AUC ratios and presented graphically. The analyses of C-peptide levels were performed using methods similar to those used to analyse the primary PD variables.

3. Results

3.1. *Study population*

The mean (range) baseline characteristics subjects (male, $n = 15$; female, $n = 4$) included in the study were as follows: age,

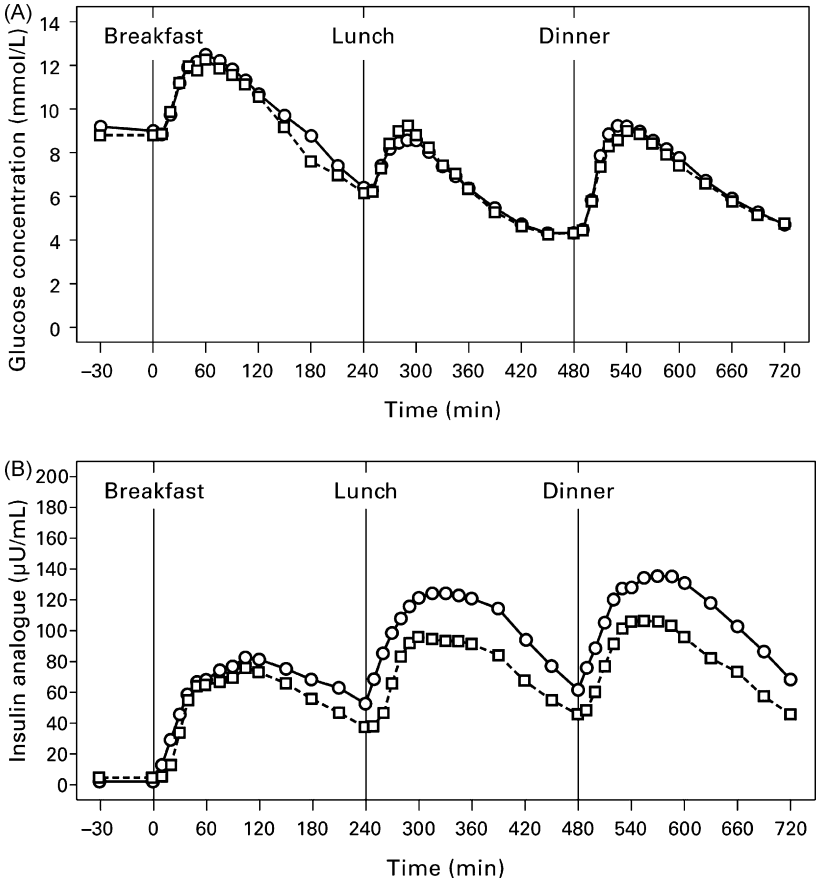

Fig. 1 – Time-concentration of (A) glucose and (B) insulin analogue profiles in obese subjects with Type 2 diabetes mellitus after treatment with 0.15 U/kg of either glulisine or lispro, immediately prior to three 500 kcal standard meals over a 12-h period (open circles = glulisine; open squares = lispro).

59.8 (41–71) years; BMI, males, 36.7 (33.2–43.8) kg/m^2; females, 40.0 (35.7–46.5) kg/m^2; HbA_{1c}, 7.8 (6.0–10.9)%.

The mean plasma glucose and serum insulin analogue concentrations are shown in Fig. 1.

3.1.1. *Plasma glucose profiles*

The mean preprandial-subtracted glucose AUC concentrations were similar on both the glulisine and lispro study days (218.94 vs. 224.32 mmol/L min, respectively).

Although there were no overall statistically significant differences in the plasma glucose profiles between glulisine and lispro (Fig. 1A and Table 1), it was evident that over the total study period (12 h), the mean of the three maximal preprandial subtracted plasma glucose concentrations (ΔGLU_{max}) was approximately 12% lower after glulisine treatment than after lispro treatment (3.55 vs. 4.06 mmol/L; $p < 0.01$). The largest between-treatment differences in ΔGLU_{max} were demonstrated during the post-lunch period where the estimated difference of ΔGLU_{max} with glulisine was approximately 25% lower compared with lispro (2.58 vs. 3.44 mmol/L; $p < 0.01$) (Fig. 2). The greatest ΔGLU_{max} (~5.0 mmol/L) occurred during the post-dinner period for both insulin analogue treatments. There were differences between GLU_{max} and GLU-T_{max} for the different meal times with the maximal glucose level being observed after breakfast ($p < 0.0001$). Mean GLU-T_{max} was lower after lunch (48 min), compared with breakfast (60 min) and dinner (62 min). There were no statistically significant between-treatment differences for GLU_{min}.

3.1.2. *Plasma insulin analogue profiles*

There was a strong linear relationship between the insulin analogue-AUC profiles (INS-AUC) and each timepoint (1, 1.5, 2 and 4 h from baseline) for both insulin analogues. However, the INS-AUC at each timepoint demonstrated that following sc injection immediately before a meal, glulisine concentrations were higher than lispro (Table 2). This was particularly notable during the post-lunch and post-dinner periods ($p < 0.01$ for all timepoints), where the estimated ratios for the two insulin analogues indicated a difference of 30–45%. Overall, the differences in INS-AUC for each insulin analogue displayed significant variation between meals ($p = 0.03$).

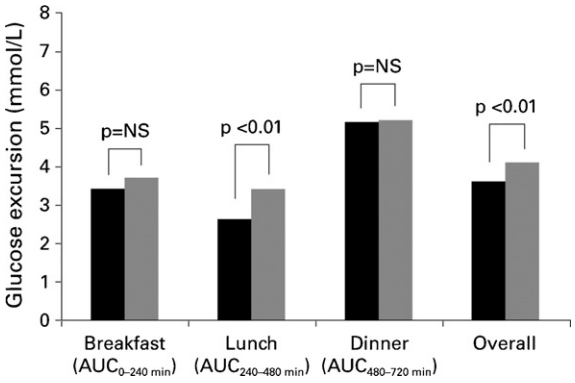

Fig. 2 – Maximum plasma glucose excursion (black bars = glulisine; grey bars = lispro). AUC = area under the curve; NS = not significant.

The INS-C_{max} following glulisine administration was significantly higher than with lispro overall (~20%; $p < 0.01$). Again, this difference was most marked during the post-lunch and post-dinner periods (Fig. 1B). In contrast, INS-T_{max} occurred ~19 min later with glulisine than with lispro ($p = 0.004$).

Analysis of the absorption rates during the first 30 min after each meal showed that there was a highly significant difference ($p < 0.0001$) between the absorption rates of the two insulin analogues, with a faster rate of absorption of glulisine (1.47 ± 0.68 μU/min) compared with lispro (0.96 ± 0.86 μU/min) during the first 30 min. The estimated difference during this period was 0.48 μU/min (90% CI: 0.31, 0.66). Overall, the estimated mean absorption rate for glulisine was 1.45 μU/min versus 0.97 μU/min for lispro.

3.1.3. *Other analyses*

The overall C-peptide levels (both C-peptide-AUC and C-peptide-C_{max}) and C-peptide-T_{max} were comparable between the two treatment arms (Table 3).

Comparison between the three meals demonstrated significant differences for C-peptide-AUC and C-peptide-C_{max} ($p < 0.0001$ for both), with the largest values for both measures recorded during the post-breakfast period. Similarly, C-peptide-T_{max} values were significantly different between

Table 1 – Pharmacodynamic (glucose) results in obese subjects with T2DM after treatment with 0.15 U/kg of either glulisine or lispro immediately prior to a 500 kcal standardised meal

Variable	Glulisine	Lispro	Ratio* (glulisine:lispro)	90% CI	*p*-Value
GLU_{max} (mmol/L)	10.00	10.25	0.98	(0.94, 1.01)	0.27
GLU_{min} (mmol/L)	4.61	4.53	1.02	(0.96, 1.07)	0.60
ΔGLU_{max} (mmol/L)					
Breakfast	3.39	3.72	0.91	(0.79, 1.04)	0.26
Lunch	2.58	3.44	0.75	(0.65, 0.86)	<0.01
Dinner	5.11	5.20	0.98	(0.86, 1.13)	0.83
Overall	3.55	4.06	0.88	(0.81, 0.95)	<0.01
GLU-T_{max} (min)	56.26	56.87	−0.60	(−5.85, 4.64)	0.85

Data = means for all variables; * = difference for GLU-T_{max}; GLU_{max} = maximum glucose concentration; GLU_{min} = minimum glucose concentration; ΔGLU_{max} = maximal glucose excursion; GLU-T_{max} = time to GLU_{max}; CI = confidence interval.

Table 2 – Pharmacokinetic (insulin analogue) results in obese subjects with T2DM after treatment with 0.15 U/kg of either glulisine or lispro immediately prior to a 500 kcal standardised meal

Variable	Meal	Time (h)	Geometric means		Ratio* (glulisine:lispro)	90% CI	*p*-Value
			Glulisine	Lispro			
INS-AUC (μU min/mL)	Breakfast	0–1	2307.37	1904.27	1.21	(1.02, 1.44)	0.07
		0–1.5	4307.24	3794.30	1.14	(0.95, 1.36)	0.23
		0–2	6653.25	5931.07	1.12	(0.97, 1.29)	0.18
		0–4	14836.81	12487.97	1.19	(1.09, 1.30)	<0.01
	Lunch	0–1	5473.18	3772.08	1.45	(1.34, 1.57)	<0.01
		0–1.5	9034.56	6537.81	1.38	(1.30, 1.47)	<0.01
		0–2	12731.39	9176.47	1.39	(1.30, 1.48)	<0.01
		0–4	23849.87	17292.60	1.38	(1.30, 1.47)	<0.01
	Dinner	0–1	5908.03	4356.89	1.36	(1.23, 1.50)	<0.01
		0–1.5	9784.40	7487.47	1.31	(1.20, 1.42)	<0.01
		0–2	13678.32	10510.95	1.30	(1.21, 1.40)	<0.01
		0–4	25767.48	18858.18	1.37	(1.30, 1.44)	<0.01
INS-C_{max} (μU/mL)	Breakfast	NA	83.86	77.35	1.08	(0.99, 1.18)	0.13
	Lunch	NA	129.01	99.92	1.29	(1.18, 1.41)	<0.01
	Dinner	NA	141.51	115.11	1.23	(1.13, 1.34)	<0.01
	Overall	NA	115.25	96.18	1.20	(1.14, 1.26)	<0.01
INS-T_{max} (min)	NA	NA	108.87	89.52	19.35	(8.45, 30.25)	<0.01

Differences between glulisine and lispro are expressed as the ratio of the geometric means, accompanied by 90% CI; * = difference for INS-T_{max}; CI = confidence interval; INS-AUC = area under the insulin analogue concentration curve; INS-C_{max} = maximum insulin analogue concentration; INS-T_{max} = time to the maximum insulin analogue concentration.

meals ($p < 0.0001$); the lowest C-peptide-T_{max} was achieved during the post-lunch period (60 min) compared with the post-breakfast and post-dinner periods (109 and 98 min, respectively).

There was some evidence of a linear relationship between glulisine:lispro ratio and skin thickness over all three meals, which was particularly notable during the post-lunch period ($r = 0.66$). Patients with skin thickness >40 mm demonstrated higher glucose disposal over the first 2 h (GIR-$AUC_{0–2\ h}$) values with glulisine versus lispro whereas the converse was true for subjects with skin thicknesses <40 mm (increased GIR-$AUC_{0–2\ h}$ values with lispro compared with glulisine). In contrast, there was no evidence of a relationship between skin thickness and the INS-AUC ratio (data not shown).

3.1.4. *Safety*

Two adverse events were reported (in two subjects), one of which was classified as serious (a lower lobe consolidation). Neither was considered to be related to the study medication. One subject demonstrated two hypoglycaemic episodes during this study, both during the lispro day, but did not require assistance. No clinically relevant changes in laboratory variables, electrocardiogram readings, vital signs, injection-site responses or physical examinations were noted.

4. Discussion

The maintenance of normoglycaemia is the primary aim of insulin replacement in patients with T2DM [6]. Improvement in glycaemic control has been shown to lower the risk of microvascular complications associated with T2DM [16–18]. The rapid-acting insulin analogues more closely resemble endogenous prandial insulin secretion compared with RHI. This study was conducted to extend our understanding of the PK and PD profile of the two rapid-acting insulin analogues, glulisine and lispro, in obese patients (BMI >30 kg/m^2) with T2DM.

The results of this study confirm previous findings in obese subjects without diabetes [15], where a faster rise in insulin concentration and faster onset of action with glulisine compared with lispro was reported. In the present study, the absorption rate of glulisine was faster during the first 30 min compared with lispro. In contrast, the plasma glucose

Table 3 – C-peptide results in obese subjects with T2DM after treatment with 0.15 U/kg of either glulisine or lispro immediately prior to a 500 kcal standardised meal

Variable	Glulisine	Lispro	Ratio* (glulisine:lispro)	90% CI	*p*-Value
C-peptide-AUC (pmol/mL min)	311.66	284.83	1.09	(0.93, 1.29)	0.35
C-peptide-C_{max} (pmol/mL)	1.82	1.74	1.05	(0.93, 1.18)	0.53
C-peptide-T_{max} (min)	86.85	91.76	−4.91	(−16.47, 6.65)	0.48

* = difference for C-peptide-T_{max}; CI = confidence interval; C-peptide-AUC = area under the C-peptide concentration curve; C-peptide-C_{max} = maximum C-peptide concentration; C-peptide-T_{max} = time to the maximum C-peptide concentration.

profiles of glulisine and lispro were similar; however, ΔGLU_{max} following every meal was significantly lower with glulisine versus lispro. The variability of blood glucose has recently been indicated in the development of long-term complications of diabetes [19]. Although the vascular damage is generally thought to be caused by the oxidative stress response to high glucose levels in complication-prone cells [19,20], postprandial glucose fluctuations exhibit a greater activation of oxidative stress compared with chronic sustained hyperglycaemia [21]. Recent evidence has emerged that the worsening of glucose homeostasis in patients with T2DM is associated with three distinct stages: a gradual loss in overall post-meal glycaemic control, followed by a deterioration during the morning period and finally by sustained hyperglycaemia during the nocturnal period [22]. Therefore, the lower postprandial blood glucose following glulisine in this group of patients may be beneficial in at least delaying the progression of T2DM, which may be explained by the characteristics of the absorption profile of glulisine, including the faster rate of absorption for glulisine compared with lispro.

The difference in absorption rates observed between glulisine and lispro may be due to the additional zinc contained in the lispro formulation, which has been shown to retard the absorption of insulin post-injection [23]. In contrast, the structural modifications made to the glulisine molecule that leave the proline at B28 unaltered mean that the glulisine molecules exist predominantly as stabilised monomers and dimers. Alteration of the proline at B28, such as in lispro, greatly reduces dimerisation of the monomer molecules, leading to unfolding of individual monomers and the subsequent development of fibril formations, which in turn are more susceptible to degradation [24,25]. Consequently, the drug formulation of lispro includes zinc to achieve sufficient shelf-life stability through the formation of stable hexameric and other higher-order aggregates [26,27]. These are less prone to solvent degradation but, when injected sc, exhibit delayed dissociation into monomers upon diffusion of the zinc ligand in the bloodstream. On the other hand, the drug formulation of glulisine contains the detergent, polysorbate 20, which reduces the unfolding of monomers but does not lead to hexamer formation and thus, theoretically, should maintain a fast rate of dissociation into monomers upon injection [24].

Whilst this study confirms the results of previous studies with respect to glucose disposal in obese patients with T2DM when glulisine is injected immediately before a meal [11] and findings in obese subjects without diabetes [15], the data also indicate that the formulation of glulisine may provide some benefits over that of lispro in terms of lower glucose excursions following a standard meal. Significantly faster absorption with higher and sustained post-meal insulin levels achieved at every meal with glulisine to that achieved with lispro, should also benefit patients who may need to cope with even larger rises in post-meal glucose concentrations. Taken together with previous data, our study not only provides evidence that glulisine has PK and PD profiles that may be of benefit to obese patients with T2DM but may also offer clinical evidence for the molecular differences between glulisine and lispro.

Preliminary results suggest that the absorption of glulisine (and lispro) is unaffected by adiposity, as reflected by the skin thickness, but the limited number of subjects in this study prevents meaningful interpretation. On the other hand, the data indicate that glulisine provides better glucose disposal (i.e. GIR-$AUC_{0-2\,h}$) in patients with skin thickness >40 mm compared with lispro. This phenomenon occurred particularly during the post-lunch period and may suggest a relationship between glucose levels and skin thickness, which could be of particular importance for obese patients. Taken together, it would be of interest to determine absorption rates in a large sample of patients with T2DM across a wider BMI range.

In conclusion, in this group of obese (BMI >30 kg/m^2) patients with T2DM, bioequivalence was demonstrated between glulisine and lispro for almost all outcomes analysed, except for ΔGLU_{max}, which was lower with glulisine versus lispro. This might be explained by a difference in the pharmacokinetics between the two insulin analogues, with higher plasma levels of insulin analogue after administration of glulisine compared with lispro. These findings may have implications in the beta-cell sparing effects of rapid-acting insulin analogues.

Acknowledgement

Editorial support was provided by the Global Publication Group of sanofi-aventis.

Conflict of interest

Stephen Luzio (Research Support) is supported by the Grant given by sanofi-aventis. Rajesh Peter, Gareth J. Dunseath and Laila Mustafa have no conflict of interest. David R. Owens (Research Support, Consultant) is supported by the Grant given by sanofi-aventis.

REFERENCES

[1] S.D. Luzio, R. Peter, G.J. Dunseath, N. Mustafa, D.R. Owens, Comparison of the pharmacodynamics and pharmacokinetics of bolus subcutaneous insulin glulisine and insulin lispro in obese patients with type 2 diabetes, Diabetologia 48 (2005) A307 (Abstract 844).
[2] S.D. Luzio, R. Peter, G.J. Dunseath, N. Mustafa, D.R. Owens, The pharmacokinetics and pharmacodynamics of insulin glulisine compared with insulin lispro in obese patients with type 2 diabetes, Diabet. Med. 23 (2006) 42 (Abstract P47).
[3] M.A. Bethel, M.N. Feinglos, Basal insulin therapy in type 2 diabetes, J. Am. Board Fam. Pract. 18 (2005) 199–204.
[4] C. Binder, T. Lauritzen, O. Faber, S. Pramming, Insulin pharmacokinetics, Diabetes Care 7 (1984) 188–199.
[5] American Diabetes Association, Insulin Administration, Diabetes Care 26 (2003) S121–S124.
[6] D.R. Owens, B. Zinman, G.B. Bolli, Insulins today and beyond, Lancet 358 (2001) 739–746.
[7] S.K. Garg, J. Rosenstock, K. Ways, Optimized Basal-bolus insulin regimens in type 1 diabetes: insulin glulisine versus regular human insulin in combination with Basal insulin glargine, Endocr. Pract. 11 (2005) 11–17.

[8] S.K. Garg, S.L. Ellis, H. Ulrich, Insulin glulisine: a new rapid-acting insulin analogue for the treatment of diabetes, Expert Opin. Pharmacother. 6 (2005) 643–651.
[9] M. Dreyer, R. Prager, A. Robinson, K. Busch, G. Ellis, E. Souhami, Efficacy and safety of insulin glulisine in patients with type 1 diabetes, Horm. Metab. Res. 37 (2005) 702–707.
[10] R.H.A. Becker, A.D. Frick, C. Kapitza, T. Heise, K. Rave, Pharmacodynamics (PD) and pharmacokinetics (PK) of insulin glulisine (GLU) compared with insulin lispro (IL) and regular human insulin (RHI) in patients with type 2 diabetes, Diabetes 53 (2004) A119 (Abstract 503-P).
[11] G. Dailey, J. Rosenstock, R.G. Moses, K. Ways, Insulin glulisine provides improved glycemic control in patients with type 2 diabetes, Diabetes Care 27 (2004) 2363–2368.
[12] H. Hauner, Managing type 2 diabetes mellitus in patients with obesity, Treat. Endocrinol. 3 (2004) 223–232.
[13] A.H. Barnett, How well do rapid-acting insulins work in obese individuals? Diabetes Obes. Metab. 8 (2006) 388–395.
[14] J.P. Vora, A. Burch, J.R. Peters, D.R. Owens, Absorption of radiolabelled soluble insulin in type 1 (insulin-dependent) diabetes: influence of subcutaneous blood flow and anthropometry, Diabet. Med. 10 (1993) 736–743.
[15] R.H. Becker, A.D. Frick, F. Burger, J.H. Potgieter, H. Scholtz, Insulin glulisine, a new rapid-acting insulin analogue, displays a rapid time-action profile in obese non-diabetic subjects, Exp. Clin. Endocrinol. Diabetes 113 (2005) 435–443.
[16] UKPDS, Intensive blood-glucose control with sulphonylureas or insulin compared with conventional treatment and risk of complications in patients with type 2 diabetes (UKPDS 33). UK Prospective Diabetes Study (UKPDS) Group, Lancet 352 (1998) 837–853.
[17] Y. Ohkubo, H. Kishikawa, E. Araki, T. Miyata, S. Isami, S. Motoyoshi, et al., Intensive insulin therapy prevents the progression of diabetic microvascular complications in Japanese patients with non-insulin-dependent diabetes mellitus: a randomized prospective 6-year study, Diabetes Res. Clin. Pract. 28 (1995) 103–117.
[18] P. Gaede, P. Vedel, H.H. Parving, O. Pedersen, Intensified multifactorial intervention in patients with type 2 diabetes mellitus and microalbuminuria: the Steno type 2 randomised study, Lancet 353 (1999) 617–622.
[19] I.B. Hirsch, M. Brownlee, Should minimal blood glucose variability become the gold standard of glycemic control? J. Diabetes Complications 19 (2005) 178–181.
[20] M. Brownlee, The pathobiology of diabetic complications: a unifying mechanism, Diabetes 54 (2005) 1615–1625.
[21] L. Monnier, E. Mas, C. Ginet, F. Michel, L. Villon, J.P. Cristol, et al., Activation of oxidative stress by acute glucose fluctuations compared with sustained chronic hyperglycemia in patients with type 2 diabetes, JAMA 295 (2006) 1681–1687.
[22] L. Monnier, C. Colette, G.J. Dunseath, D.R. Owens, The loss of postprandial glycemic control precedes stepwise deterioration of fasting with worsening diabetes, Diabetes Care 30 (2007) 263–269.
[23] R.H. Rosskamp, G. Park, Long-acting insulin analogs, Diabetes Care 22 (1999) B109–B113.
[24] R.H. Becker, Insulin glulisine complementing basal insulins: a review of structure and activity, Diabetes Technol. Ther. 9 (2007) 109–121.
[25] V. Sluzky, J.A. Tamada, A.M. Klibanov, R. Langer, Kinetics of insulin aggregation in aqueous solutions upon agitation in the presence of hydrophobic surfaces, Proc. Natl. Acad. Sci. U.S.A. 88 (1991) 9377–9381.
[26] D.L. Bakaysa, J. Radziuk, H.A. Havel, M.L. Brader, S. Li, S.W. Dodd, et al., Physicochemical basis for the rapid time-action of LysB28ProB29-insulin: dissociation of a protein-ligand complex, Protein Sci. 5 (1996) 2521–2531.
[27] S. Bhatnagar, D. Srivastava, M.S. Jayadev, A.K. Dubey, Molecular variants and derivatives of insulin for improved glycemic control in diabetes, Prog. Biophys. Mol. Biol. 91 (2006) 199–228.