Electronic supplementary material

Clinical Pharmacokinetics

Clinical Pharmacology of Fast-Acting Insulin Aspart Versus Insulin Aspart Measured as Free or Total Insulin Aspart and the Relation to Anti-Insulin Aspart Antibody Levels in Subjects with Type 1 Diabetes Mellitus

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Table S1 Trial information

Trial no.	Reference	Trial registration no.	N	Key inclusion criteria			
				BMI (kg/m^2)	HbA_{1c}		C-peptide (nmol/L)
					(%)	(mmol/mol)	
1	Heise et al. [1]	NCT01618188	51	18.0-28.0	≤8.5%	69	≤0.3
2	Heise et al. [2]	NCT02003677	44[a]	18.5-28.0	≤9.5%	80	≤0.3
3	Shiramoto et al. [3]	NCT01934712	40	18.5-28.0	≤9.0%	75	≤0.3
4	Basu et al. [4]	NCT02568280	40	18.5-28.0	≤8.5%	69	≤0.3
5	Russell-Jones et al. [5]	NCT01831765	1040	≤35.0	7.0-9.5%	53-80	ND

BMI, body mass index; HbA_{1c}, glycosylated haemoglobin; N, number of subjects; ND, not determined
[a] Only those subjects where total insulin aspart was measured (i.e. the first 44 randomised subjects) are included for Trial 2

Table S2 Pharmacokinetic blood sampling scheme in the clinical pharmacology trials

Nominal time	Trial 1	Trial 2	Trial 3	Trial 4
00:00[a]	X	X	X	X
00:02	X	X	X	
00:04	X	X	X	
00:05				X
00:06	X	X	X	
00:08	X	X	X	
00:10	X	X	X	X
00:12	X	X	X	
00:14	X	X	X	
00:15				X
00:16	X	X	X	
00:18	X	X	X	
00:20	X	X	X	X
00:25	X	X	X	X
00:30	X	X	X	X
00:35	X	X	X	
00:40	X	X	X	X
00:45	X	X	X	
00:50	X	X	X	X
00:55	X	X	X	
01:00	X	X	X	X
01:05	X	X	X	
01:10	X	X	X	
01:15	X	X	X	X
01:20	X	X	X	
01:30	X	X	X	X
01:40	X	X	X	
01:50	X	X	X	
02:00	X	X	X	X
02:15	X	X	X	
02:30	X	X	X	X
02:45	X	X	X	
03:00	X	X	X	X
03:30	X	X	X	X
04:00	X	X	X	X
05:00	X	X	X	X
06:00	X	X	X	X
07:00	X	X	X	X
08:00	X	X	X	X
10:00	X	X	X	
12:00	X	X	X	

[a] Within 5 minutes pre-dose in Trial 1, within 2 minutes pre-dose in Trials 2 and 4 and within 10 minutes pre-dose in Trial 3

Table S3 Baseline characteristics

Characteristic		Clinical pharmacology trials		Phase IIIa trial
		Pooled PK[a]	Pooled PD[b]	Trial 5
Males/females	N	113/62	82/30	616/424
Age, years	Mean	42	42	45
	Min	21	21	18
	Max	73	73	83
Body weight, kg	Mean	72.0	71.4	80.0
	Min	48.6	48.6	45.3
	Max	100.0	97.4	140.0
BMI, kg/m^2	Mean	23.9	23.7	26.7
	Min	18.5	18.5	17.0
	Max	28.7	28.7	37.9
Duration of diabetes, years	Mean	21	21	20
	Min	1	1	1
	Max	57	57	65
Baseline HbA_{1c}, %	Mean	7.3	7.4	7.6
	Min	5.2	5.2	5.6
	Max	9.2	8.9	9.8
Baseline HbA_{1c}, mmol/mol	Mean	52	52	59
	Min	30	30	38
	Max	71	68	84
Anti-IAsp antibodies, %B/T[c]	Mean	10.79	10.98	18.0
	Min	-0.11	-0.11	0.2
	Max	66.94	66.94	86.7
Fasting C-peptide, nmol/L	Mean	0.03	0.04	ND
	Min	0.00	0.01	ND
	Max	0.30	0.30	ND

BMI, body mass index; HbA_{1c}, glycosylated haemoglobin; IAsp, insulin aspart; Max, maximum; Min, minimum; N, number of subjects; ND, not determined; PK, pharmacokinetics; PD, pharmacodynamics

[a] Trials 1 to 4 (i.e. the 4 clinical pharmacology trials)

[b] Trials 1 to 3 (i.e. those 3 clinical pharmacology trials using glucose clamp to assess pharmacodynamics)

[c] Pre-dose in Trials 1 to 4 and at 26 weeks of treatment in Trial 5

Table S4 Onset of exposure for faster aspart and insulin aspart after subcutaneous administration in subjects with type 1 diabetes mellitus when using an assay to measure free or total insulin aspart

Variable	Free IAsp				Total IAsp			
	LS mean Faster aspart	LS mean IAsp	Treatment difference Faster aspart - IAsp [95% CI]	*p* value[a]	LS mean Faster aspart	LS mean IAsp	Treatment difference Faster aspart - IAsp [95% CI]	*p* value[a]
Onset of appearance (min)	3.4	7.5	-4.1 [-4.7;-3.6]	<0.001	2.7	5.2	-2.5 [-2.8;-2.1]	<0.001
$t_{Early\ 50\ \%\ Cmax}$ (min)[b]	17.5	26.2	-8.8 [-10.0;-7.5]	<0.001	17.8	25.4	-7.6 [-8.8;-6.4]	<0.001
t_{max} (min)	55.2	66.0	-10.8 [-15.4;-6.3]	<0.001	54.2	64.7	-10.6 [-14.7;-6.5]	<0.001

Results are based on 175 individual pharmacokinetic profiles per treatment
CI, confidence interval; IAsp, insulin aspart; LS mean, least square mean; $t_{Early\ 50\ \%\ Cmax}$, time to 50% of maximum IAsp concentration in the early part of the pharmacokinetic profile; t_{max}, time to maximum IAsp concentration
[a] For treatment comparison of faster aspart vs. IAsp
[b] Not determined in Trial 4

Table S5 Early exposure for faster aspart and insulin aspart after subcutaneous administration in subjects with type 1 diabetes mellitus when using an assay to measure free or total insulin aspart

Variable	Free IAsp				Total IAsp			
	LS mean Faster aspart	LS mean IAsp	Treatment ratio Faster aspart/IAsp [95% CI]	*p* value[a]	LS mean Faster aspart	LS mean IAsp	Treatment ratio Faster aspart/IAsp [95% CI]	*p* value[a]
$AUC_{IAsp,0\text{-}15\ min}$ (pmol·h/L)	16.4	4.8	3.40 [3.00;3.85]	<0.001	25.9	9.0	2.87 [2.58;3.20]	<0.001
$AUC_{IAsp,0\text{-}30\ min}$ (pmol·h/L)	67.7	36.0	1.88 [1.74;2.04]	<0.001	117.3	66.5	1.77 [1.64;1.90]	<0.001
$AUC_{IAsp,0\text{-}60\ min}$ (pmol·h/L)	211.1	164.1	1.29 [1.22;1.36]	<0.001	372.2	295.0	1.26 [1.20;1.33]	<0.001

Results are based on 175 individual pharmacokinetic profiles per treatment. The dose was adjusted to 0.2 U/kg in all subjects
$AUC_{IAsp,0\text{-}x\ min}$, area under the concentration-time curve for IAsp from time zero to x min; CI, confidence interval; IAsp, insulin aspart; LS mean, least square mean
[a] For treatment comparison of faster aspart vs. IAsp

Table S6 Offset of exposure for faster aspart and insulin aspart after subcutaneous administration in subjects with type 1 diabetes mellitus when using an assay to measure free or total insulin aspart

Variable	Free IAsp				Total IAsp			
	LS mean Faster aspart	LS mean IAsp	Treatment difference Faster aspart - IAsp [95% CI]	*p* value[a]	LS mean Faster aspart	LS mean IAsp	Treatment difference Faster aspart - IAsp [95% CI]	*p* value[a]
$t_{Late\ 50\ \%\ Cmax}$ (min)	143	157	-13.8 [-20.0;-7.6]	<0.001	140	154	-14.0 [-20.1;-7.9]	<0.001
Variable	Free IAsp				Total IAsp			
	LS mean Faster aspart	LS mean IAsp	Treatment ratio Faster aspart/IAsp [95% CI]	*p* value[a]	LS mean Faster aspart	LS mean IAsp	Treatment ratio Faster aspart/IAsp [95% CI]	*p* value[a]
$AUC_{IAsp,2\text{-}t}$ (pmol·h/L)	232	262	0.88 [0.81;0.96]	0.005	428	479	0.89 [0.84;0.96]	0.001

Results are based on 175 individual pharmacokinetic profiles per treatment. For $AUC_{IAsp,2\text{-}t}$, the dose was adjusted to 0.2 U/kg in all subjects.
$AUC_{IAsp,2\text{-}t}$, area under the concentration-time curve for IAsp from 2 h; CI, confidence interval; IAsp, insulin aspart; LS mean, least square mean; $t_{Late\ 50\ \%\ Cmax}$, time to 50% of maximum IAsp concentration in the late part of the pharmacokinetic profile
[a] For treatment comparison of faster aspart vs. IAsp

Table S7 Overall exposure for faster aspart and insulin aspart after subcutaneous administration in subjects with type 1 diabetes mellitus when using an assay to measure free or total insulin aspart

Variable	Free IAsp				Total IAsp			
	LS mean Faster aspart	LS mean IAsp	Treatment ratio Faster aspart/IAsp [95% CI]	*p* value[a]	LS mean Faster aspart	LS mean IAsp	Treatment ratio Faster aspart/IAsp [95% CI]	*p* value[a]
$AUC_{IAsp,0\text{-}t}$ (pmol·h/L)	719.5	720.1	1.00 [0.97;1.03]	0.962	1292.7	1285.9	1.01 [0.97;1.04]	0.760
$C_{max,IAsp}$ (pmol/L)	330.8	318.2	1.04 [0.99;1.09]	0.092	572.9	553.1	1.04 [0.99;1.08]	0.101

Results are based on 175 individual pharmacokinetic profiles per treatment. The dose was adjusted to 0.2 U/kg in all subjects
$AUC_{IAsp,0\text{-}t}$, total area under the concentration-time curve for IAsp; CI, confidence interval; C_{max}, maximum IAsp concentration; IAsp, insulin aspart; LS mean, least square mean
[a] For treatment comparison of faster aspart vs. IAsp

Prediction of $AUC_{IAsp,0\text{-}t}$ in the phase IIIa trial

In the phase IIIa trial (Trial 5), $AUC_{IAsp,0\text{-}t}$ was predicted from the two IAsp concentration measurements done 1 and 2 hours post-dose at 26 weeks of treatment (C_1 and C_2, respectively). The prediction was based on data from available clinical pharmacology trials with faster aspart and IAsp (101 pharmacokinetic profiles to predict free IAsp for mealtime dosing arms, 600 pharmacokinetic profiles to predict free IAsp for post-meal dosing arms and 189 pharmacokinetic profiles to predict total IAsp). The prediction was done using multiple linear regression without intercept for the relationship between IAsp concentrations at 1 and 2 hours post-dose and $AUC_{IAsp,0\text{-}t}$. The equations used to predict $AUC_{IAsp,0\text{-}t}$ are shown in Table S8. R^2 (i.e. the fraction of variation explained by the linear model) was between 0.51 and 0.84 for the different predictions.

Table S8 Equations used for the prediction of $AUC_{IAsp,0\text{-}t}$ in the phase IIIa trial

	Free IAsp		Total IAsp	
	Both measurements available	Only 2-hour measurement available[a]	Both measurements available	Only 2-hour measurement available[a]
Mealtime dosing arms[b]	$0.66 \cdot C_1 + 2.67 \cdot C_2$	$3.55 \cdot C_2$	$0.54 \cdot C_1 + 3.08 \cdot C_2$	$3.77 \cdot C_2$
Post-meal dosing arm[c]	$0.38 \cdot C_1 + 2.58 \cdot C_2$	$3.00 \cdot C_2$	$0.014 \cdot C_1 + 3.31 \cdot C_2$	$3.33 \cdot C_2$

AUC, area under the curve; C_1 and C_2, free or total insulin aspart concentrations at 1 and 2 hours post-dose; IAsp, insulin aspart

[a] If only the 1-hour measurement was available $AUC_{IAsp,0\text{-}t}$ was not predicted

[b] Both faster aspart and IAsp

[c] The actual time points of insulin aspart measurement were 40 minutes post-dose for C_1 and 100 minutes post-dose for C_2

References

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