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ORIGINAL ARTICLE

Effect of initial combination therapy with sitagliptin and metformin on β-cell function in patients with type 2 diabetes

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Aim: To examine the effect of sitagliptin and metformin, alone and in combination, on modelled parameters of β-cell function in patients with type 2 diabetes.

Methods: The data used in the present analyses are from a 104-week study, which included a 24-week, placebo- and active controlled phase followed by a 30-week, active controlled, continuation phase and an additional 50-week, active controlled extension phase. Patients were randomised to one of six blinded treatments: sitagliptin 50 mg + metformin 1000 mg b.i.d., sitagliptin 50 mg + metformin 500 mg b.i.d., metformin 1000 mg b.i.d., metformin 500 mg b.i.d., sitagliptin 100 mg q.d. or placebo. Patients on placebo were switched in a blinded manner to metformin 1000 mg b.i.d. at week 24. Subsets of patients volunteered to undergo frequently sampled meal tolerance tests at baseline and at weeks 24, 54 and 104. β-cell responsivity was assessed with the C-peptide minimal model. The static component (Φ_s) estimates the rate of insulin secretion related to above-basal glucose concentration. The dynamic component (Φ_d) is related to the rate of change in glucose. The total index (Φ_{total}) represents the overall response to a glycaemic stimulus and is calculated as a function of Φ_s and Φ_d. Insulin sensitivity was estimated with the Matsuda index (ISI). The disposition index, which assesses insulin secretion relative to the prevailing insulin sensitivity, was calculated based on the Φ_{total} and ISI.

Results: At week 24, substantial reductions in postmeal glucose were observed with all active treatment groups relative to the placebo group. Φ_s, Φ_{total} and the disposition index were significantly improved from baseline at week 24 with all active treatments relative to placebo. Generally larger effects were observed with the initial combination of sitagliptin and metformin relative to the monotherapy groups. When expressed as median percent change from baseline, Φ_s increased from baseline by 137 and 177% in the low- and high-dose combination groups and by 85, 54, 73 and −9% in the high-dose metformin, low-dose metformin, sitagliptin monotherapy and placebo groups, respectively. At weeks 54 and 104, the combination treatment groups continued to demonstrate greater improvements in β-cell function relative to their respective monotherapy groups.

Conclusions: After 24 weeks of therapy, relative to placebo, initial treatment with sitagliptin or metformin monotherapy improved β-cell function; moreover, initial combination therapy demonstrated larger improvements than the individual monotherapies. Improvements in β-cell function were found with treatments for up to 2 years.

Keywords: β-cell, DPP-IV inhibitor, metformin, type 2 diabetes

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Introduction

β-cell dysfunction (i.e. inadequate insulin secretion) and insulin resistance (i.e. inadequate insulin action) are key pathologic defects in type 2 diabetes [1]. Despite continued treatment, β-cell function deteriorates over time in patients with type 2 diabetes, highlighting the progressive nature of this disease [2,3]. Furthermore, insulin resistance increases over time regardless of baseline glucose tolerance category [4]. A reciprocal relationship exists whereby reduced insulin sensitivity leads to a compensatory increase in insulin secretion and vice versa under normal conditions. This islet adaptation is integral to normal glucose homeostasis, and inadequate changes in adaptation contribute to the development or worsening of type 2 diabetes [5,6]. Treatments that target both β-cell dysfunction and insulin resistance may be effective in preventing or slowing the progression of type 2 diabetes.

The incretins, glucagon-like peptide-1 (GLP-1) and glucose-dependent insulinotropic polypeptide (GIP), improve β-cell mass, morphology and function *in vitro* and in animal models [7]. Therapeutic agents, such as GLP-1 agonists and dipeptidyl peptidase-4 (DPP-4) inhibitors, target the incretin pathway [8]. In clinical trials, the DPP-4 inhibitor, sitagliptin, improved fasting and postprandial glycaemic control and measures of β-cell function in patients with type 2 diabetes, with minimal effects on measures of insulin resistance/sensitivity [9,10]. Metformin has been found to increase

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GLP-1 levels in humans [11,12]. In healthy and type 2 diabetic subjects, co-administration of sitagliptin and metformin produced approximately additive enhancement of active GLP-1 levels relative to the individual agents [11,13]. In addition, metformin increases insulin sensitivity in patients with type 2 diabetes [3]. When administered as initial combination therapy, sitagliptin and metformin substantially improved glycaemic control and fasting measures of homeostatic model assessment β-cell function (i.e. HOMA-β, proinsulin/insulin ratio) and homeostatic model assessment insulin resistance (HOMA-IR) over 2 years [14–16].

Fasting measures of β-cell function and insulin action may not adequately reflect the glucose-dependent actions of sitagliptin in the postprandial state. β-cell function can also be assessed in the postprandial state with a C-peptide-based model, which quantifies the overall amount of insulin secretion and partitions these effects into static (i.e. response to a given glucose concentration) and dynamic (i.e. response to a change in glucose) components [17]. Further, a disposition index can be calculated to estimate insulin secretion in the context of insulin action. Therefore, it was of interest to determine in patients with type 2 diabetes whether initial combination therapy with sitagliptin and metformin enhanced β-cell function, as assessed by a C-peptide model, and to evaluate these effects over 24 weeks and in patients who completed treatment out to 104 weeks.

Methods

The design of the clinical study and the efficacy and safety findings over 104 weeks have been published elsewhere [14–16]. Briefly, patients with type 2 diabetes who provided informed consent were randomised to one of six treatment groups for 24 weeks: sitagliptin 50 mg + metformin 1000 mg b.i.d. (high-dose combination), sitagliptin 50 mg + metformin 500 mg b.i.d. (low-dose combination), metformin 1000 mg b.i.d., metformin 500 mg b.i.d., sitagliptin 100 mg q.d. or placebo. At week 24, patients randomised to active treatment continued their treatment for an additional 30 weeks (54 weeks total) and those randomised to placebo were switched to metformin 1000 mg b.i.d. At week 54, patients were required to reconsent to extend their participation and treatment for an additional 50 weeks (104 weeks total). Patients and investigators remained blinded to treatment assignment throughout the 104 weeks. Glycaemic rescue medication was used throughout the study for patients who met progressively stricter glycaemic criteria [14–16].

Of the 1091 patients randomised in this trial, a subset of patients volunteered to undergo frequently sampled meal tolerance tests at baseline (prior to first dose of study medication) and at weeks 24, 54 and 104 (30 min after taking morning dose of study medication). For patients who met the glycaemic rescue criteria or discontinued during the study, a frequently sampled meal tolerance test was completed, if appropriate, prior to receiving rescue medication or discontinuation. The meal challenge was a mixed meal consisting of a nutrition bar and drink (approximately 460 kcal total; 75 g of carbohydrate, 9 g of fat and 18 g of protein). Patients were instructed to consume the entire meal within 15 min. Blood samples for the meal tolerance tests were collected at the following time points relative to the start of the meal: −35, −10, 0 (immediately prior to the meal), 10, 20, 30, 60, 90, 120 and 180 min. Plasma glucose and serum C-peptide and insulin concentrations were assayed at a central laboratory (PPD Global Central Labs, LLC, Highland Heights, KY and Zaventem, Belgium).

Using the C-peptide minimal model [17], β-cell function was assessed from glucose and C-peptide concentrations obtained during the frequently sampled meal tolerance tests. The model assumes that insulin secretion is made up of three components: static, dynamic and basal. The static component (Φ_s) estimates the provision of new insulin to the releasable pool and provides an assessment of the rate of insulin secretion related to above-basal glucose concentration. The dynamic component (Φ_d) represents secretion of promptly releasable insulin and is related to the rate of increase in glucose. The basal sensitivity index (Φ_b) is a measure of β-cell responsivity to glucose under basal conditions. The total responsivity index (Φ_{total}) is a pooled parameter, defined as the average insulin secretion rate above the basal level over the average glucose concentration, calculated as a function of Φ_s and Φ_d. The Matsuda index (ISI) was used to estimate insulin sensitivity [18]. Disposition index characterises insulin secretion in the context of insulin action and is the product of Φ_{total} and ISI.

Statistical Analyses

Data were included in the analysis if proper procedures for the meal tolerance test were followed and results were available at baseline and the postrandomisation time point of interest (i.e. week 24, 54 or 104). The following rules were implemented to ensure consistency in handling blinded data during the data modelling process. Both the glucose and C-peptide data were required to have a basal value and values for all postzero time points during a given meal test to allow for good precision in the parameter estimation. The basal value was calculated as the mean of the available values from the −35, −10 and 0 min point. Following randomisation, missing values were imputed with the last observation carried forward approach within each time period (i.e. 0–24 weeks, 24–54 weeks or 54–104 weeks), but were not carried forward from one time period to the subsequent time period. The indices of β-cell function from the C-peptide minimal model together with their precision were estimated by nonlinear least squares (NLS) using the SAAM II software [19].

The primary analysis included patients who underwent frequently sampled meal tolerance tests at baseline and week 24 and had evaluable C-peptide modelling data at both time points. For the responses to the meal tolerance tests (e.g. glucose area under the time-concentration curve (AUC), insulin AUC, C-peptide AUC, ratio of insulin AUC/glucose AUC), an analysis of covariance (ANCOVA) model compared treatment groups for continuous efficacy parameters, focusing on change from baseline at week 24, with baseline values and prior antihyperglycaemic agent use as covariates. The between-group differences (active vs. placebo) for these endpoints were estimated by evaluating the placebo-adjusted least squares (LS) mean change from baseline and 95% confidence interval (CI).

Table 1. Baseline characteristics of patients who underwent a frequently sampled meal tolerance test at baseline and week 24, week 54 or week 104.

Parameter	Placebo*	Sitagliptin 100 mg q.d.	Metformin 500 mg b.i.d.	Metformin 1000 mg b.i.d.	Sitagliptin 50 mg + MET 500 mg b.i.d.	Sitagliptin 50 mg + MET 1000 mg b.i.d.
Cohort through week 24						
n	45	55	49	59	52	46
Age (years)	53.9 ± 12.3	51.8 ± 9.8	53.8 ± 10.8	53.8 ± 9.6	54.4 ± 10.0	54.7 ± 8.4
Male, n (%)	21 (47)	26 (47)	28 (57)	30 (51)	32 (62)	22 (48)
BMI (kg/m^2)	33.5 ± 7.5	31.7 ± 5.7	30.9 ± 6.0	32.8 ± 7.8	32.0 ± 6.5	33.2 ± 7.5
HbA1c (%)	8.8 ± 1.1	8.8 ± 1.0	8.7 ± 0.9	8.5 ± 0.8	8.6 ± 0.9	8.8 ± 1.0
FPG (mg/dl)	198.4 ± 55.0	198.4 ± 53.0	206.4 ± 49.9	197.3 ± 48.0	195.8 ± 53.5	204.8 ± 48.8
Duration of T2DM (years)	4.8 ± 5.0	4.5 ± 5.1	4.2 ± 3.3	4.2 ± 4.1	4.3 ± 3.6	5.3 ± 4.3
Cohort through week 54						
n	—	33	26	46	37	44
Age (years)	—	53.9 ± 9.7	52.5 ± 10.8	54.6 ± 9.0	55.3 ± 10.0	54.4 ± 8.7
Male, n (%)	—	16 (49)	14 (54)	22 (48)	20 (54)	20 (46)
BMI (kg/m^2)	—	31.1 ± 4.9	32.3 ± 6.1	32.9 ± 7.7	31.1 ± 6.6	33.2 ± 7.2
HbA1c (%)	—	8.5 ± 0.9	8.5 ± 0.7	8.4 ± 0.9	8.6 ± 0.9	8.7 ± 0.9
FPG (mg/dl)	—	177.8 ± 42.5	191.9 ± 46.9	193.1 ± 44.9	189.3 ± 46.9	198.2 ± 49.0
Duration of T2DM (years)	—	5.1 ± 5.7	4.4 ± 4.2	3.7 ± 3.8	4.6 ± 4.4	5.1 ± 4.5
Cohort through week 104						
n	—	13	23	26	29	29
Age (years)	—	53.9 ± 9.2	55.7 ± 9.6	56.2 ± 9.4	57.7 ± 7.4	56.8 ± 7.2
Male, n (%)	—	7 (54)	11 (48)	11 (42)	14 (48)	13 (45)
BMI (kg/m^2)	—	31.0 ± 5.7	30.6 ± 6.1	30.3 ± 5.0	30.1 ± 7.1	32.0 ± 6.2
HbA1c (%)	—	8.1 ± 0.9	8.4 ± 0.7	8.5 ± 0.8	8.5 ± 0.8	8.8 ± 1.0
FPG (mg/dl)	—	165.2 ± 39.7	182.9 ± 44.2	189.3 ± 47.2	177.9 ± 40.7	206.4 ± 48.5
Duration of T2DM (years)	—	5.4 ± 7.4	4.6 ± 4.3	4.0 ± 3.5	4.1 ± 4.6	6.1 ± 4.9

Data are expressed as mean ± standard deviation or frequency [n (%)]. MET, metformin; BMI, body mass index; FPG, fasting plasma glucose; T2DM, type 2 diabetes mellitus; HbAlc, haemoglobin A1c.

*Patients who were randomised to placebo and completed treatment through week 24 were switched to metformin 1000 mg b.i.d. for the remainder of the study. Results from patients in this group were not included in the analyses after week 24.

Since the data for the β-cell indices were not normally distributed, the analyses focused on median change from baseline. Hodges–Lehman estimates of median placebo-adjusted treatment effects and their 95% CI based on the Wilcoxon's rank sum test were calculated with the 24-week data [20]. P-values for between-group comparisons were obtained using the above ANCOVA model, substituting the change from baseline at week 24 and the baseline value with the corresponding Tukey's normalised ranks [21]. Similar analyses were used to evaluate β-cell modelling results (Φ_s) for weeks 54 and 104, except only within-treatment group differences from baseline were calculated. Partial Spearman's rank-order correlation coefficients were calculated to assess the relationship between the changes from baseline at week 24 in β-cell function-related parameters and glycaemic efficacy [i.e. haemoglobin A1c (HbA1c)], controlled for the effect of treatment. A p-value <0.05 was considered statistically significant.

Results

Patient Characteristics

Week 24. A subset of 500 patients volunteered to undergo frequently sampled meal tolerance tests. Of these 500 patients, 306 had evaluable data to assess glucose, insulin and C-peptide responses at baseline and week 24. Baseline demographics and disease characteristics of these patients were similar across groups (Table 1), and were generally similar to the randomised cohort [14], which suggests that no bias was introduced by analysing data in a subset of the randomised population. For the β-cell modelling analyses (e.g. Φ_s results), 294 patients had evaluable data. Patients (n = 206) were excluded from the β-cell modelling analyses for missing data (91%), physiologically implausible data (5%), or not consuming the entire meal (4%) at baseline, while on treatment, or both.

Glucose, Insulin and C-peptide Responses to Meal Tolerance Tests

Consistent with the previously published findings for the three-point meal tests (2-h postprandial glucose, 2-h glucose AUC values) from the randomised cohort [14], all active therapies led to significant reductions in postprandial glucose (3-h glucose AUC) during the 10-point meal test compared to placebo at week 24, with the largest reduction observed in the high-dose combination group (figure 1 and Table 2). In the setting of substantial reductions in postprandial glucose, increases in postprandial 3-h insulin and C-peptide AUC were observed in all active treatment groups relative to placebo. These increases were non-significant except for insulin AUC in the low-dose combination group (Table 2). The ratio of insulin AUC/glucose AUC increased in all active treatment groups, with significant differences from placebo observed in the high-dose metformin and in both combination treatment groups (Table 2).

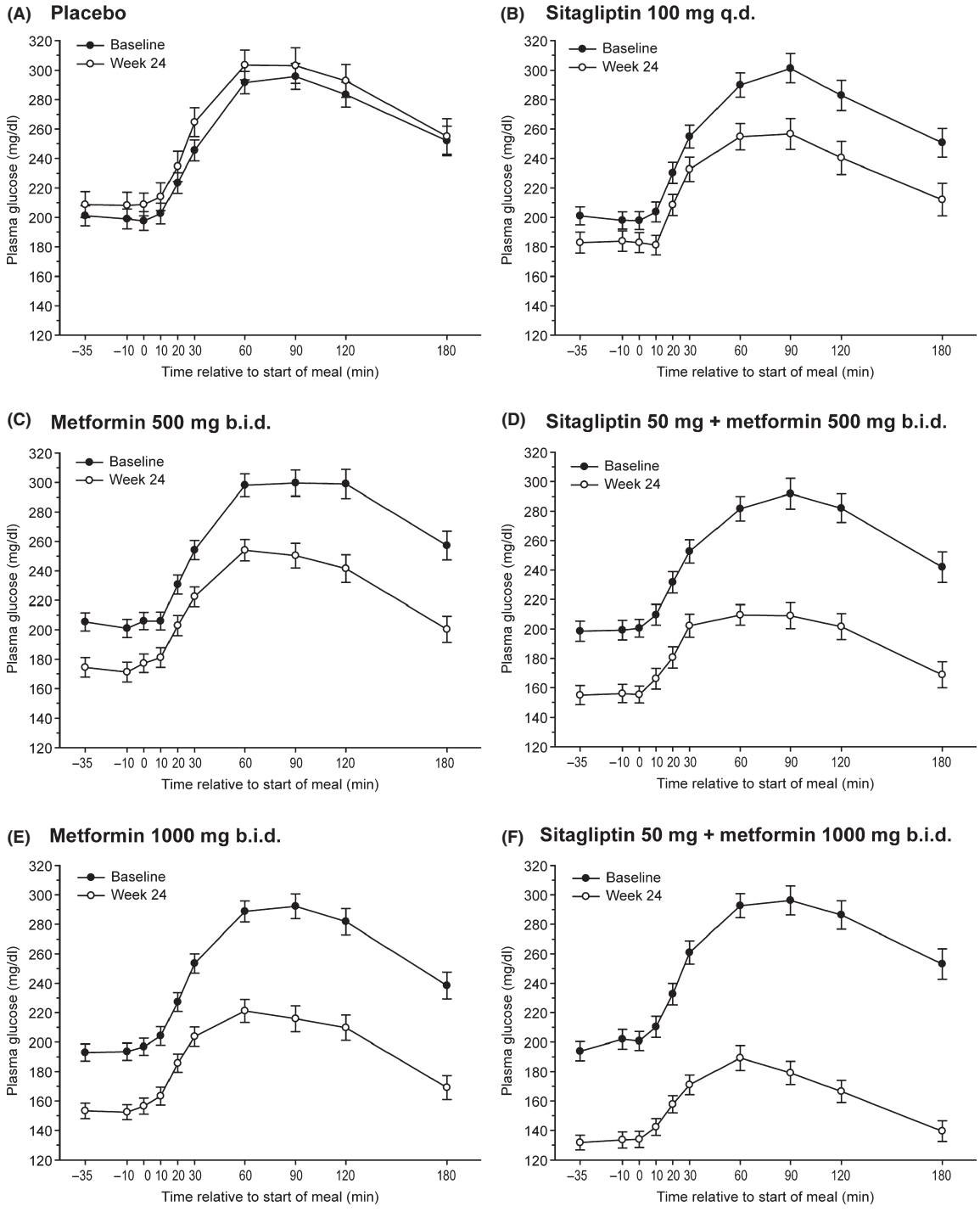

Figure 1. Baseline and week 24 plasma glucose response during a frequently sampled meal tolerance test for each treatment.

β-cell Responsivity (Φ), Insulin Sensitivity Index and Disposition Index

Following a mixed meal, parameters of β-cell responsiveness, Φ_s and Φ_{total}, significantly improved with all active treatments compared with placebo, with the largest effects observed in the combination groups relative to their respective monotherapy groups at week 24 (Table 3). The effect of treatments on Φ_s is also presented graphically in figure 2. Steady-state insulin secretion in response to plasma glucose (slope of line, Φ_s) increased with all active treatments relative to baseline, whereas it was unchanged with placebo (figure 2). When expressed as median percent change from baseline, Φ_s increased from baseline by 137 and 177% in the low-dose and high-dose combination groups and by 54, 85 and 73% in the low-dose metformin, high-dose metformin and sitagliptin monotherapy groups,

Table 2. Glucose, insulin and C-peptide responses to frequently sampled meal tolerance tests at baseline and week 24.

Parameter	Placebo	Sitagliptin 100 mg q.d.	Metformin 500 mg b.i.d.	Metformin 1000 mg b.i.d.	Sitagliptin 50 mg + MET 500 mg b.i.d.	Sitagliptin 50 mg + MET 1000 mg b.i.d.
Glucose 3-h AUC (mg·h/dl), n	45	55	49	59	52	46
Baseline	798 ± 197	806 ± 204	834 ± 190	797 ± 179	778 ± 199	828 ± 198
Change from baseline	33 (−12, 78)	−88 (−129, −47)	−136 (−180, −93)	−205 (−244, −166)	−233 (−275, −190)	−325 (−370, −280)
Change from placebo	—	−121 (−182, −60)**	−169 (−232, −106)**	−238 (−298, −178)**	−265 (−327, −204)**	−358 (−422, −294)**
Insulin 3-h AUC (μIU·h/ml), n	44	46	43	50	43	40
Baseline	132 ± 94	101 ± 65	121 ± 95	131 ± 87	127 ± 100	115 ± 54
Change from baseline	−5 (−20, 10)	7 (−7.5, 21.4)	5 (−10, 20)	13 (−1, 27)	20 (5, 34)	4 (−11, 20)
Change from placebo	—	12 (−9, 33)	10 (−11, 31)	18 (−2, 38)	25 (4, 45)*	10 (−12, 31)
C-peptide 3-h AUC (ng·h/ml), n	45	55	49	59	52	46
Baseline	17.3 ± 7.1	15.8 ± 6.2	16.0 ± 6.5	18.4 ± 6.9	16.4 ± 6.6	17.6 ± 6.6
Change from baseline	−0.3 (−1.4, 0.8)	1.1 (0.1, 2.1)	0.4 (−0.7, 1.4)	0.5 (−0.5, 1.4)	0.8 (−0.2, 1.9)	0.8 (−0.3, 1.9)
Change from placebo	—	1.3 (−0.1, −2.8)	0.6 (−0.9, 1.2)	0.8 (−0.7, 2.2)	1.1 (−0.4, 2.6)	1.0 (−0.5, 2.6)
Insulin AUC/glucose AUC ratio, n	44	46	43	50	43	40
Baseline	0.18 ± 0.17	0.14 ± 0.10	0.17 ± 0.16	0.18 ± 0.15	0.18 ± 0.17	0.16 ± 0.10
Change from baseline	−0.01 (−0.04, 0.03)	0.03 (0.00, 0.06)	0.04 (0.01, 0.07)	0.08 (0.05, 0.11)	0.10 (0.07, 0.14)	0.10 (0.06, 0.13)
Change from placebo	—	0.04 (−0.01, 0.08)	0.04 (−0.00, 0.09)	0.09 (0.04, 0.13)**	0.11 (0.06, 0.16)**	0.11 (0.06, 0.15)**

Baseline and week 24 data are expressed as mean ± standard deviation. Change from baseline and placebo data are expressed as within-group and between-group least square (LS) mean change from baseline [95% confidence interval (CI)], respectively. MET, metformin; AUC, area under the concentration-time curve.
*$p \leq 0.05$ versus placebo; **$p \leq 0.001$ versus placebo.

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Table 3. β-cell modelling results from frequently sampled meal tolerance tests administered at baseline and week 24.

Parameter	Placebo	Sitagliptin 100 mg q.d.	Metformin 500 mg b.i.d.	Metformin 1000 mg b.i.d.	Sitagliptin 50 mg + MET 500 mg b.i.d.	Sitagliptin 50 mg + MET 1000 mg b.i.d.
Φ_s, 10^{-9}/min, n	44	52	46	58	50	44
Baseline	15.5	13.7	12.2	15.5	15.7	14.4
Change from baseline	−1.3 (−4.5, 1.9)	10.6 (6.5, 14.6)	5.9 (2.3, 9.5)	13.0 (8.5, 17.5)	18.4 (12.1, 24.7)	18.9 (11.8, 26.1)
Change from placebo	—	11.2 (6.5, 16.0)**	6.7 (2.5, 11.2)*	13.5 (8.1, 19.2)**	19.4 (13.2, 26.7)**	20.1 (14.4, 27.1)**
Φ_d, 10^{-9}, n	45	52	48	58	51	45
Baseline	460.9	404.7	417.6	479.3	481.1	455.6
Change from baseline	5.2 (−66.4, 76.7)	24.2 (−76.9, 127.9)	31.4 (−39.9, 102.7)	54.1 (−23.2, 131.3)	21.8 (−75.9, 119.5)	132.1 (2.3, 261.9)
Change from placebo	—	37.0 (−73.3, 154.9)	11.7 (−103.7, 100.4)	70.7 (−30.3, 184.5)	26.9 (−85.3, 153.2)	151.0 (17.6, 296.4)*
Φ_b, 10^{-9}/min, n	44	52	46	58	50	44
Baseline	5.4	5.3	4.6	5.5	4.7	5.4
Change from baseline	0.2 (−0.3, 0.7)	0.6 (0.0, 1.1)	0.5 (0.1, 0.9)	1.3 (0.8, 1.8)	1.6 (1.1, 2.0)	2.2 (1.5, 3.0)
Change from placebo	—	0.8 (0.2, 1.4)*	0.7 (0.1, 1.2)	1.3 (0.7, 2.0)**	1.7 (1.0, 2.3)**	2.1 (1.3, 3.0)**
Φ_{total}, 10^{-9}/min, n	44	50	45	57	49	43
Baseline	8.5	7.7	6.4	7.4	6.9	7.6
Change from baseline	−0.8 (−1.7, 0.0)	1.4 (0.6, 2.2)	1.2 (0.4, 2.0)	2.4 (1.7, 3.0)	2.2 (1.1, 3.3)	3.5 (2.0, 5.1)
Change from placebo	—	2.2 (1.3, 3.3)**	2.0 (0.9, 3.0)*	2.9 (1.9, 4.0)**	3.1 (1.8, 4.4)**	4.5 (3.0, 6.1)**
ISI, n	50	49	48	52	54	42
Baseline	2.7	3.4	2.8	2.1	2.5	2.1
Change from baseline	−0.2 (−0.6, 0.1)	0.1 (−0.4, 0.5)	0.1 (−0.4, 0.6)	0.7 (0.2, 1.2)	0.6 (0.1, 1.1)	1.1 (0.6, 1.7)
Change from placebo	—	0.3 (−0.3, 0.8)	0.5 (−0.0, 1.1)	1.1 (0.6, 1.7)**	0.9 (0.4, 1.6)**	1.4 (0.9, 2.1)**
Disposition index, n	40	40	38	48	42	35
Baseline	17.6	18.8	17.4	20.0	17.4	17.0
Change from baseline	−3.4 (−5.9, −0.9)	2.2 (−3.0, 7.4)	5.2 (−0.8, 11.1)	11.6 (6.6, 16.7)	10.6 (3.7, 17.5)	20.0 (11.5, 28.5)
Change from placebo	—	7.0 (2.1, 13.1)*	10.0 (4.0, 16.8)*	15.0 (9.7, 20.9)**	15.8 (8.6, 23.8)**	27.0 (19.6, 36.3)**

Baseline data are expressed as median. Change from baseline or placebo data are expressed as median change [95% confidence interval (CI) for median]. MET, metformin, ISI, insulin sensitivity index.
*$p \leq 0.05$ versus placebo; **$p \leq 0.001$ versus placebo.

respectively. The median percent change from baseline in Φ_s was −9% for the placebo group. For Φ_b, significant increases relative to placebo were observed in all active treatment groups, except in the low-dose metformin group. For Φ_d, a significant increase was found in the high-dose combination group, whereas small numerical increases were observed in the other groups compared with placebo (Table 3).

Insulin sensitivity, using the Matsuda index, increased in all active treatment groups, with larger and statistically significant differences from placebo observed in the high-dose metformin and combination treatment groups (Table 3). The disposition index significantly increased in all active treatment groups relative to placebo, with the largest effects observed in the combination groups relative to their respective monotherapy groups (Table 3).

The relationship between changes from baseline in β-cell function parameters and HbA1c was assessed with partial Spearman's rank-order correlation coefficients. Modest, significant inverse correlations ($\rho = -0.3$ to -0.4; $p < 0.001$) were found between changes in β-cell function parameters and glycaemic efficacy (Table 4). A small, but significant correlation was found between changes in insulin sensitivity and glycaemic efficacy (Table 4). In addition, there was a positive and significant relationship between the change from baseline in HOMA-β, a fasting measure of β-cell function, and modelled parameters of β-cell responsivity [Φ_{total} ($\rho = 0.51$; $p < 0.001$); Φ_s ($\rho = 0.24$; $p < 0.001$)].

Weeks 54 and 104. The number of patients who completed the frequently sampled meal tests decreased over time due to patients receiving rescue medication, discontinuing the study or not completing the tests for a variety of reasons. Given the progressively (over time) stricter rescue criteria for inadequate glycaemic control and the differences in glycaemic efficacy between the combination and monotherapy groups, more patients in the combination groups relative to the monotherapy groups completed frequently sampled meal tolerance tests at the later time points. Because of these factors, it was not unexpected that the patients who completed meal tests at weeks 54 and 104 generally had lower mean HbA1c and fasting plasma glucose (FPG) values at baseline relative to those who completed the meal test at week 24 (Table 1). For the median change from baseline in Φ_s, the trends appeared to be similar within groups over 54 and 104 weeks, with larger changes observed in the combination groups relative to the monotherapy groups (figure 3). When results for HOMA-β were evaluated over time, similar trends within groups were observed (figure 4).

Discussion

In the present study, using the C-peptide minimal model [17], the responsiveness (Φ_s) of the β-cell to glucose significantly improved in all active treatment regimens relative to placebo over 24 weeks, with numerically greater increases observed in the sitagliptin and metformin combination groups (increased up to 177% over baseline value) relative to the respective

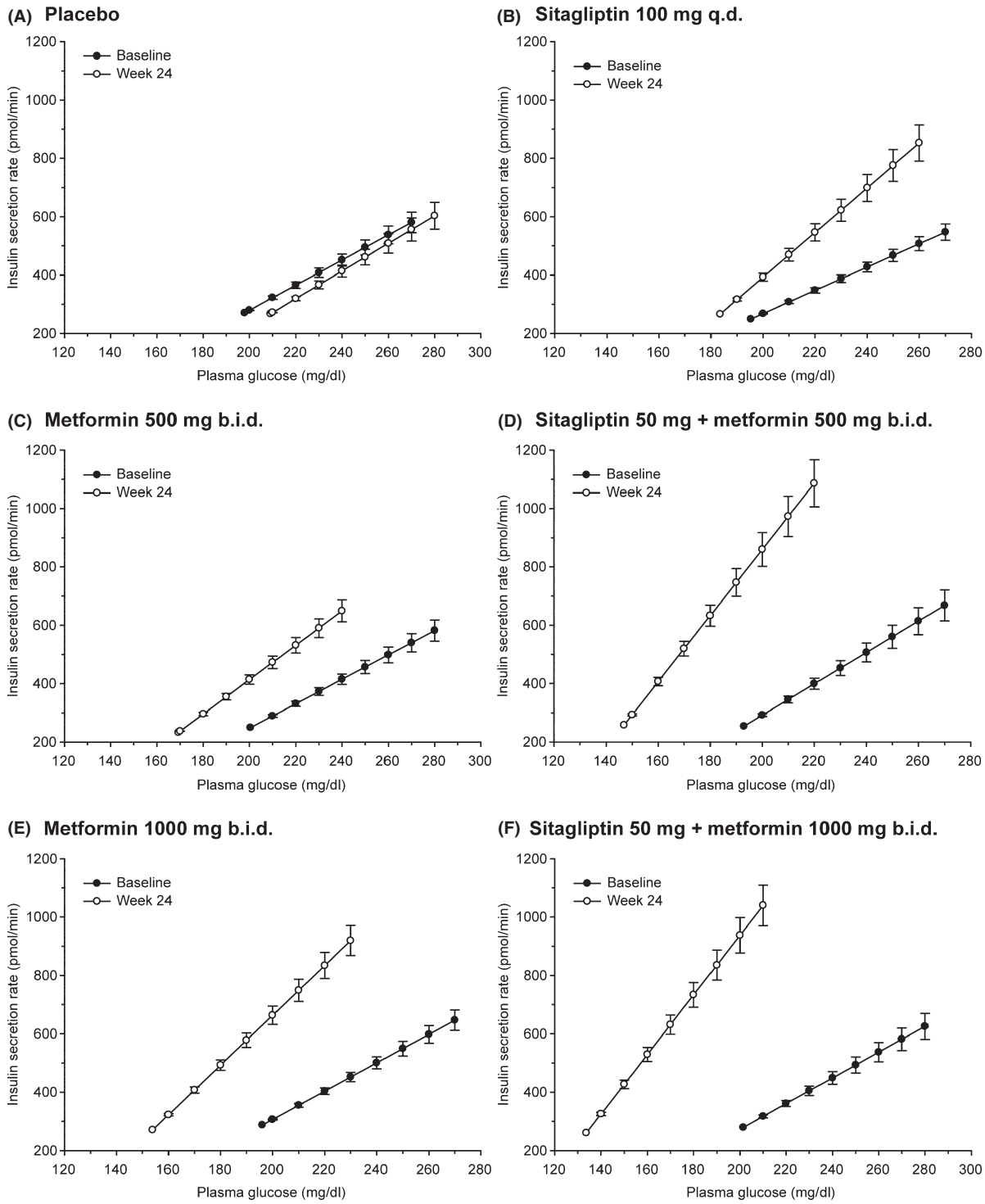

Figure 2. Steady-state rate of insulin secretion expressed as a function of glucose concentration (i.e. Φ_s) at baseline and week 24 for each treatment group.

monotherapy groups (up to 85%). There were numeric, but not significant (except in the high-dose combination group), improvements in the dynamic response to glucose (Φ_d), which is related to an increased rate of docking and exocytosis of insulin-containing granules. Consistent with the present findings, Campioni et al. [22] demonstrated that the incretin effect increases insulin secretion through greater static responses than dynamic responses. In this study, when the static and dynamic responses were assessed together, the overall responsiveness (i.e. Φ_{total}) of the β-cell was significantly increased in all active treatment groups at week 24. The increases in the ratio of insulin AUC/glucose AUC support the

Table 4. Partial Spearman's rank-order correlation coefficients for the relationship between changes from baseline in β-cell function-related parameters and in haemoglobin A1c (HbA1c) at week 24, controlled for treatment group.

β-cell function parameter	Δ HbA1c ρ	p-Value
$\Delta\Phi_s$	−0.31	<0.001
$\Delta\Phi_b$	−0.40	<0.001
$\Delta\Phi_{total}$	−0.38	<0.001
Δ Insulin sensitivity index	−0.14	0.013
Δ Disposition index	−0.39	<0.001

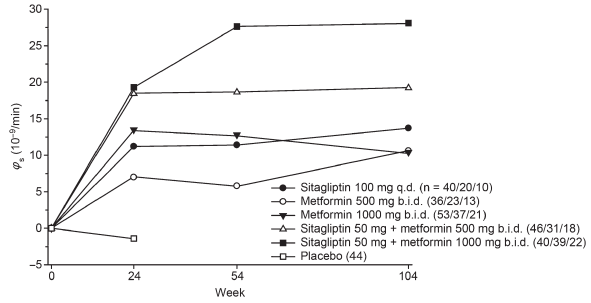

Figure 3. Median change from baseline in Φ_s for the cohorts of patients who had baseline and measurements at each time point. Sample sizes at each time point are noted within the figure.

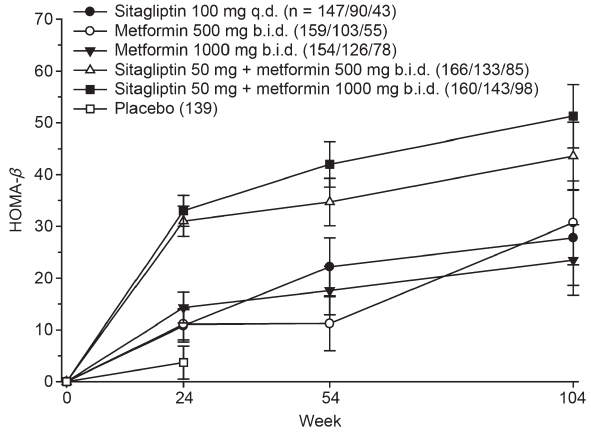

Figure 4. Least squares (LS) mean change (SE) from baseline in HOMA-β for the specific efficacy population at each time point. Sample sizes at each time point are noted within the figure.

improved β-cell response found with the active treatments at week 24. These findings are also consistent with the previously published results from the present study in which fasting measures of β-cell function, HOMA-β and the proinsulin/insulin ratio were significantly improved with initial combination therapy relative to the monotherapy groups over 24 weeks [14]. Further, similar model-based results were observed when sitagliptin was used as monotherapy or as add-on therapy to metformin for up to 24 weeks [23,24]. Overall, initial treatment of type 2 diabetes with sitagliptin and metformin alone or in combination improved multiple measures of β-cell function, with greater effects observed with the combined agents.

Incretins enhance pancreatic β-cell function in various animal and cell culture models [7]. Sitagliptin increases active GLP-1 and GIP levels in healthy subjects and patients with type 2 diabetes [25]. Metformin increased total GLP-1 levels, and when co-administered with sitagliptin, this combination produced approximately additive effects on active GLP-1 levels in healthy subjects and patients with type 2 diabetes [11,13]. Thus, the greater improvements in model-based parameters of β-cell function in this study are consistent with the effects of sitagliptin and metformin on incretins.

In addition to the changes in β-cell responsiveness, insulin sensitivity increased with the initial combination of sitagliptin and metformin. The effect on ISI is consistent with significant changes in HOMA-IR previously reported with this combination [14]. The improvement in insulin sensitivity is driven primarily by metformin, as sitagliptin has not been shown to influence parameters of insulin resistance/sensitivity [9]. The interplay between insulin secretion and insulin sensitivity is paramount for maintaining or normalizing glucose homeostasis [1]. Under normal conditions, changes in insulin sensitivity are compensated by inverse changes in β-cell responsiveness such that the product of insulin secretion and insulin sensitivity, the disposition index, remains constant [26,27]. This relationship between insulin secretion and insulin sensitivity is best described with a hyperbolic curve. Individuals who are able to maintain normal glucose tolerance in response to a decrease in insulin sensitivity have a constant disposition index (or remain on the curve) due to the compensatory response in insulin secretion (i.e. islet adaptation). In contrast, patients with a deteriorating glycaemic control experience a leftward shift below the hyperbolic curve [6,26,27]. In the present study, the changes in insulin secretion and insulin sensitivity with the initial combination of sitagliptin and metformin resulted in significant improvements in the disposition index after 24 weeks, suggesting a rightward shift towards the normal hyperbolic curve for these patients.

Treatment with sitagliptin and metformin alone or in combination led to significant reductions in postprandial glycaemic excursions following a meal in the present study. Improvements in parameters of β-cell function (i.e. Φ_s and Φ_{total}) may contribute to these marked reductions, as demonstrated by the significant inverse correlations between the change from baseline in these parameters and the change in HbA1c at week 24. In addition, the disposition index, as a function of β-cell function and insulin sensitivity, provides a measure of the ability to respond to hyperglycaemic challenges. This is demonstrated in the present study with the relationship ($\rho = -0.39$) between the change in disposition index and the change in HbA1c from baseline at week 24. A similar correlation was reported with another DPP-4 inhibitor [28]. A cause and effect relationship cannot be definitively established, however, as reversal of glucose toxicity may have contributed to the positive changes observed in β-cell function [29]. Overall, glycaemic control and β-cell function improved with sitagliptin and with metformin, with greater effects observed with the agents combined.

Because type 2 diabetes is a progressive disease with a continuing decline in β-cell function, it is of interest to evaluate the effect of treatment on β-cell function over time. In the present study, modelled β-cell function was determined in subsets of patients who participated in the extended meal tolerance at baseline and at weeks 24, 54 or 104. The improvements in β-cell function, Φ_s or HOMA-β, were observed for up to 104 weeks. However, it is not possible to ascertain whether the effects of these agents on β-cell function could be considered as disease modifying or as simply a reflection of continued favourable glycaemic effects. Previous studies with the DPP-4 inhibitor, vildagliptin, found that the improvements in β-cell function noted with treatment up to 1 year were not sustained following a 4-week washout period [30]. However, washout periods without maintenance of glycaemic control by another means (e.g. insulin therapy) are potentially confounded by the glucotoxicity resulting from abrupt discontinuation of antihyperglycaemic therapy. Such glucotoxicity may obscure assessment of any long-term β-cell benefit afforded by a preceding therapy. Despite this, following 2 years of treatment and a 4 to 7-day washout period, the addition of sitagliptin to ongoing metformin led to better maintenance of β-cell function relative to baseline compared with the addition of glipizide in patients with type 2 diabetes and inadequate glycaemic control on metformin monotherapy [31]. However, because β-cell function in this study was not measured prior to the washout after the completion of the 2-year treatment period, it is not possible to understand any detrimental impact of the washout period in this study [31].

The following limitations should be considered when interpreting these results. The patients volunteered and were not randomised to undergo the frequently sampled meal tolerance tests. This may introduce some selection bias although the patients who underwent the frequently sampled meal tolerance tests had baseline characteristics similar to the overall randomised population. The number of patients who completed the frequently sampled meal tolerance tests declined over the 2-year study. Numerous reasons impacted the sample size over time: progressively stricter glycaemic control criteria, which led to patients receiving glycaemic rescue medication or discontinuing the study, patients withdrawing or discontinuing the study for other reasons, and patients withdrawing consent for or missing frequently sampled meal tolerance tests at the follow-up time points. Therefore, the results at 54 and 104 weeks should be interpreted with caution although they appear to follow the trends observed at 24 weeks.

In summary, the initial combination of sitagliptin and metformin enhanced the responsiveness of pancreatic β-cells to glucose in both the fasting and postprandial states at 24 weeks in patients with type 2 diabetes. The improvement in β-cell function appeared to be maintained over the 2-year treatment period.

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Conflict of Interest

D. W-H., L. X., R. T., G. T. G., J. J., K. D. K. and B. G. were involved in the concept and design of the study and in the data collection and/or analysis. All authors were involved in interpretation of the results. M. J. D. drafted the article and all authors were involved in the critical revisions, discussions and approval of the article. All authors are employees of Merck Sharp & Dohme Corp., the manufacturer of sitagliptin.

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