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BRIEF REPORT

Pioglitazone prevents the increase in plasma ketone concentration associated with dapagliflozin in insulin-treated T2DM patients: Results from the Qatar Study

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Because of the unique mechanism of action of sodium-glucose co-transport inhibitors (SGLT2i), which is independent of insulin secretion and insulin action, members of this class of drugs effectively lower plasma glucose concentration when used in combination with all other antidiabetic agents, including insulin. Increased plasma ketone concentration has been reported in association with SGLT2i initiation, which, under certain clinical conditions, has developed into diabetic ketoacidosis. The daily insulin dose often is reduced at the time of initiating SGLT2i therapy in insulin-treated patients to avoid hypoglycaemia. However, reduction of insulin dose can increase the risk of ketoacidosis. In the present study, we examined the effect of the addition of dapagliflozin plus pioglitazone on plasma ketone concentration in insulin-treated T2DM patients and compared the results to the effect of dapagliflozin alone. A total of 18 poorly controlled, insulin-treated T2DM participants in the Qatar Study received dapagliflozin (10 mg) plus pioglitazone (30 mg), and 10 poorly controlled non-insulin-treated T2DM patients received dapagliflozin (10 mg) alone for 4 months. Dapagliflozin plus pioglitazone produced a robust decrease in HbA1c (−1.4%) and resulted in a 50% reduction in daily insulin dose, from 133 to 66 units, while dapagliflozin alone caused a 0.8% reduction in HbA1c. Dapagliflozin caused a four-fold increase in fasting plasma ketone concentration, while the combination of pioglitazone plus dapagliflozin was not associated with a significant increase (0.13 vs 0.15 mM) in plasma ketone concentration or in risk of hypoglycaemia. These results demonstrate that the addition of pioglitazone to dapagliflozin prevents the increase in plasma ketone concentration associated with SGLT2i therapy.

KEYWORDS

dapagliflozin, insulin therapy, pioglitazone, Qatar study, T2DM

1 | INTRODUCTION

We[1–4] and others[5,6] have shown that, in addition to lowering plasma glucose concentration, SGLT2i exert multiple other metabolic actions in T2DM patients including improved insulin sensitivity, enhanced beta cell function, and increased plasma free fatty acid (FFA) concentration, total body fat oxidation and ketone production. Although, in absolute terms, the increase in plasma ketone concentration is small, from approximately 0.2 mM to approximately 0.4 mM, it occurs in the vast majority (>90%) of patients receiving SGLT2i therapy,[7,8], and in 10% to 20% of patients the plasma ketone level exceeds the upper limit of the normal range.[9–11] Because of the reduction in daily insulin dose, the increase in plasma ketone concentration associated with the initiation of SGLT2i therapy was highest in patients receiving insulin therapy.

Under certain clinical conditions, acute illness for example, SGLT2i therapy has led to the development of clinically significant

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diabetic ketoacidosis despite a normal/near normal plasma glucose concentration, that is, euglycaemic ketoacidosis.[7] We have shown previously that the magnitude of increase in the plasma ketone concentration following empagliflozin administration is strongly and inversely associated with the magnitude of decrease in plasma glucose and insulin concentrations and is strongly correlated with the increase in plasma FFA concentration.[8] Because pioglitazone is a strong inhibitor of lipolysis and lowers plasma FFA concentration independent of plasma insulin concentration,[12] we hypothesized that the combination of pioglitazone and SGLT2i will prevent the increase in plasma ketone concentration. The present study was designed to test this hypothesis in insulin-treated T2DM patients who participated in the Qatar Study.

2 | RESEARCH DESIGN AND METHODS

The Qatar Study[13] is a prospective, randomized, open label study that examined the efficacy of basal/bolus insulin versus combination therapy with pioglitazone plus exenatide in poorly controlled T2DM patients who were receiving maximal doses of metformin plus sulfonylurea. Patients in the insulin arm who failed to achieve the treatment goal of HbA1c < 7.0% as the result of inability to escalate the insulin dose because of hypoglycaemic events received rescue therapy. A total of 18 therapy-failure patients (Table 1) in the insulin-treated arm received a combination of dapagliflozin (10 mg) and pioglitazone (30 mg) as rescue therapy. Both agents were initiated simultaneously and plasma glucose concentrations were monitored daily in the morning and before meals. Patients were seen weekly, and basal and preprandial insulin doses were adjusted according to home-measured blood glucose levels to avoid hypoglycaemia. Because patients had HbA1c > 7.0%, no change in insulin dose was made at the time of intiating rescue therapy. Patients were instructed to return to the clinic with home-measured blood glucose levels within 1 week, or sooner, if hypoglycaemic events occurred, and adjustment in daily insulin dose was made based upon the results of home-measured blood glucose values. The basal insulin dose was reduced to eliminate fasting hypoglycaemia and the prandial insulin dose was reduced to eliminate postprandial hypoglycaemic events. Clinical and anthropometric parameters were collected. HbA1c, fasting plasma C-peptide concentration, lipid profile and plasma ketone concentration were measured before and 4 months after initiation of rescue therapy.

TABLE 1 Clinical and metabolic characteristics of study participants before and 4 months after initiation of rescue therapy with pioglitazone plus dapagliflozin

	Baseline	Four months	*P* value
Number	18		
Age (y)	53 ± 2		
Gender (f/m)	9/9		
Diabetes duration (y)	11.2 ± 0.5		
BMI (kg/m^2)	36.4 ± 1.7	36.1 ± 1.8	0.71
Weight (kg)	101 ± 4	100 ± 5	0.69
BP (mm Hg)	128/72	124/69	0.41
Insulin dose (unit/d)	133 ± 16	66 ± 15	<0.0001
Haematocrit (%)	38.9 ± 0.7	41.2 ± 1.1	0.009
Total cholesterol (mM)	3.7 ± 0.2	3.8 ± 0.1	0.71
Triglycerides (mM)	1.7 ± 0.2	1.3 ± 0.2	0.25
HDL-C (mM)	1.04 ± 0.06	1.16 ± 0.06	0.02
FPG (mg/dL)	136 ± 8	102 ± 4	<0.0001
HbA1c (%)	8.3 ± 0.2%	6.9 ± 0.2%	<0.0001
Fasting plasma C-peptide (ng/mL)	2.1 ± 0.3	1.3 ± 0.1	0.008
Fasting plasma ketone (mM)	0.13 ± 0.01	0.15 ± 0.01	0.26

Abbreviations: BMI, body mass index; BP, blood pressure (systolic/diastolic); HDL, high density lipoprotein; FPG, fasting plasma glucose; HbA1c, glycated haemoglobin.

Because no participant in the Qatar Study received therapy with dapagliflozin alone, and to compare the effect of combination therapy with pioglitazone plus dapagliflozin on plasma ketone concentration to that of dapagliflozin therapy alone, we included a second cohort of 10 poorly controlled T2DM patients (age, 51 ± 2 years; BMI, 31.2 ± 1.9 kg/m^2; diabetes duration, 7.6 ± 2.0 years; HbA1c, 8.5 ± 0.4% and fasting plasma ketones, 0.05 ± 0.01 mM) who were treated with maximal doses of metformin plus glipizide. Other than diabetes, participants were in good general health based upon medical history, physical examination, CBC and blood chemistries. Participants received dapagliflozin (10 mg/d) every morning in addition to metformin and glipizide for 4 months. Fasting plasma glucose, C-peptide and ketones concentrations and HbA1c were measured monthly in all patients.

2.1 | Statistical analysis

Data are presented as mean ± SEM. A paired t-test was utilized to compare the effect of pioglitazone plus dapagliflozin on various parameters. The increase in plasma ketone concentration caused by dapagliflozin was compared to that caused by dapagliflozin plus pioglitazone with two-way ANOVA.

3 | RESULTS

Participants (n = 18) in the Qatar Study were middle age, obese and had long-standing diabetes (Table 1). Despite a large dose of insulin (mean daily dose, 133 ± 16 units per day; glargine, 87 units and aspart, 46 units), participants had fasting plasma glucose of 136 ± 8 mg/dL and HbA1c of 8.3 ± 0.2%. At 4 months after initiation of rescue therapy with dapagliflozin plus pioglitazone, the insulin dose had been reduced by 50% (total of 66 units; glargine, 55 units and aspart, 11 units) and the fasting plasma C-peptide concentration was decreased by 38%, indicating a significant decrease in endogenous insulin secretion as well. Despite the marked decrease in both exogenous insulin and endogenous insulin secretion, both the fasting plasma glucose concentration and HbA1c decreased markedly (Table 1). Despite the marked decrease in both plasma glucose concentration and insulin dose, there was no change in the fasting plasma ketone concentration (0.13 ± 0.01 vs 0.15 ± 0.01 mM; *P* = NS) after treatment with dapagliflozin plus pioglitazone. Five patients experienced a small increase in plasma ketone concentration to levels within the normal range (Figure 1A), while plasma ketone concentration decreased

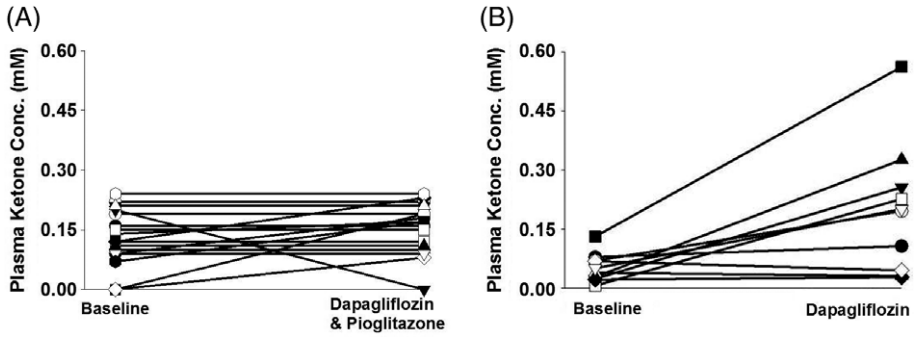

FIGURE 1 Plasma ketone concentration at baseline and after treatment with pioglitazone plus dapagliflozin A, or dapagliflozin alone, B in T2DM patients

to zero in one patient, and remained unchanged in the remaining 12 patients. No patient experienced an increase in fasting plasma ketone concentration above the normal level or developed diabetic ketoacidosis after treatment with pioglitazone plus dapagliflozin.

There was no significant change in body weight at 4 months after initiation of therapy with pioglitazone plus dapagliflozin (Table 1). One patient experienced ankle oedema, which was managed with diuretic therapy. After initiating rescue therapy with pioglitazone plus dapagliflozin, seven patients experienced hypoglycaemia (FPG < 60 mg/dL) which subsided after down titration of the insulin dose. In two patients, insulin therapy was discontinued.

In patients receiving dapagliflozin (10 mg/d) alone, HbA1c decreased by 0.8% (from 8.5 ± 0.4 to 7.7 ± 0.3%; $P < 0.001$), accompanied by a four-fold increase in plasma ketone concentration (from 0.05 ± 0.01 to 0.20 ± 0.06 mM; $P = 0.03$), and in two out of 10 patients the fasting plasma ketone concentration increased to above normal level (mean = 0.44 mM; n = 2) (Figure 1B). No change was made in background antihyperglycaemic therapy during the study in patients receiving dapagliflozin alone. Further, there was a small non-significant increase in fasting plasma C-peptide concentration (4.3 ± 0.5 vs 5.1 ± 0.6 ng/mL; $P = 0.19$). The absolute increase in plasma ketone concentration caused by dapagliflozin (+0.15 ± 0.02 mM) was significantly greater than that caused by dapagliflozin plus pioglitazone (+0.03 ± 0.02 mM, $P = 0.02$).

4 | DISCUSSION

The major finding of the present study is that pioglitazone may be a useful adjunctive therapy to prevent the increase in plasma ketone concentration caused by dapagliflozin therapy in T2DM patients. Despite the marked reduction in insulin dose, by 50%, the decrease in endogenous insulin secretion, by 38%, and the decrease in plasma glucose concentration, by 34 mg/dL, there was no significant increase in plasma ketone concentration in patients receiving pioglitazone plus dapagliflozin, demonstrating that the addition of pioglitazone to an SGLT2i could prevent an increase in plasma ketone concentration. This was in marked contrast to the effect of treatment with dapagliflozin alone (Figure 1B), which caused a four-fold increase in fasting plasma ketone concentration that rose above the upper limit of the normal level in 20% of patients. The impact of pioglitazone therapy in blocking the increase in plasma ketone concentration caused by dapaglfilozin therapy is more striking considering that the insulin dose was reduced by 50% in patients receiving pioglitazone plus dapagliflozin, whereas the background therapy remained unchanged in patients receiving dapagliflozin alone. Moreover, dapagliflozin caused a small non-significant increase in fasting plasma C-peptide concentration, suggesting that endogenous insulin secretion was not decreased or was increased slightly. Nonetheless, dapagliflozin therapy alone caused a four-fold increase in fasting plasma ketone concentration in this group. Further, only a relatively small number of patients (n = 18) was required to demonstrate the significant impact of pioglitazone therapy in inhibiting the rise in plasma ketone concentration caused by dapagliflozin therapy. It could be argued that the presence of background therapy with insulin in the group receiving pioglitazone plus dapagliflozin ameliorated the increase in plasma ketone concentration caused by SGLT2i. No previous study has examined the impact of background insulin therapy on the increase in plasma ketone concentration caused by SGLT2i. However, the majority of T2DM patients with reported diabetic ketoacidosis attributed to SGLT2i use had received insulin as part of their background antidiabetic treatment.[9,10] In one case series, 59% of T2DM patients who developed diabetic ketoacidosis (n = 34) were receiving two or more daily insulin injections.[10] Moreover, 36% of T2DM patients (n = 640) reported to the adverse event centre of the US Food and Drug Administration[14] as having diabetic ketoacidosis as the result of SGLT2i use were receiving insulin as part of their background therapy, suggesting that the presence of insulin in background antidiabetic therapy does not reduce the risk of diabetic ketoacidosis associated with SGLT2i use. Collectively, the present results suggest that pioglitazone inhibits the increase in ketone production caused by SGLT2i, and we hypothesize that combination therapy with pioglitazone plus SGLT2i will reduce the risk of ketoacidosis in T2DM patients.[7] Consistent with this hypothesis, only four of 64 (6.25%) patients who were receiving pioglitazone as part of their background therapy were reported to have developed ketoacidosis following SGLT2i administration[7,9–11] compared to 36% of insulin-treated T2DM patients.[14] A larger study is warranted to examine whether the addition of pioglitazone to dapagliflozin therapy prevents the SGLT2i-induced increase in plasma ketone concentration and the development of euglycaemic ketoacidosis reported with SGLT2i therapy.[7,9–11] We did not measure plasma-free

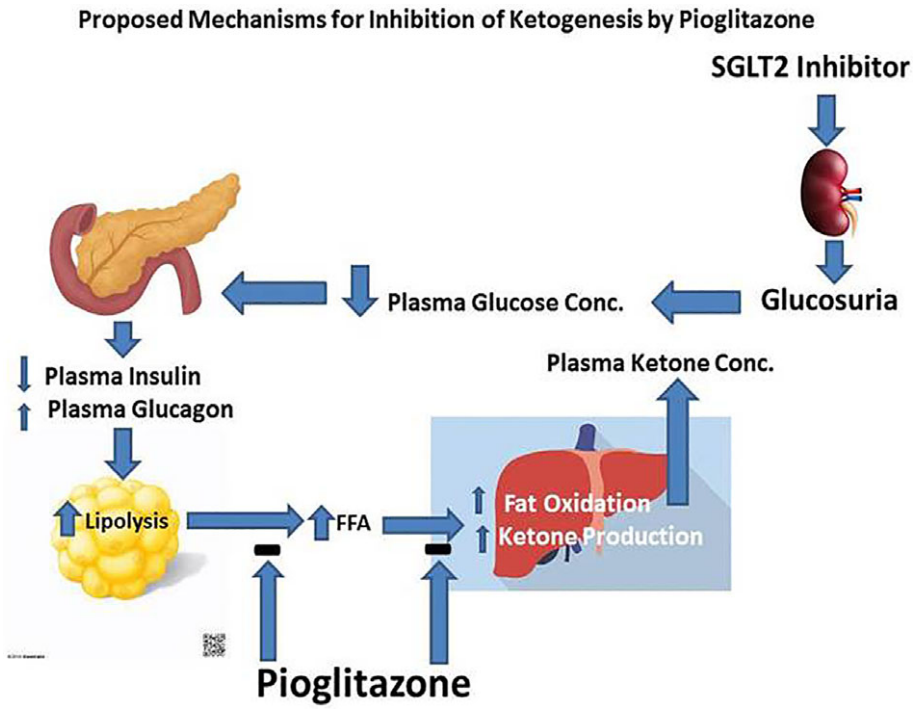

FIGURE 2 Proposed mechanisms by which pioglitazone inhibits the increase in plasma ketone concentration caused by SGLT2 inhibition

fatty acid concentration in the present study. Thus, we could hypothesize only concerning the potential mechanism(s) by which pioglitazone prevents an increase in plasma ketone concentration (Figure 2). Previous studies from our group[2,8] and others[6] have demonstrated an increase in plasma FFA concentration and total body fat oxidation following initiation of SGLT2i therapy.

The results of the present study also demonstrate that the initiation of combination therapy with pioglitazone plus dapagliflozin is very effective in lowering the plasma glucose concentration in insulin-treated patients who fail to achieve the treatment goal of HbA1c < 7.0%. Despite a 50% reduction in daily insulin dose and a significant reduction in endogenous insulin secretion, as evident by a 38% reduction in fasting plasma C-peptide concentration, combination therapy with dapagliflozin plus pioglitazone reduced HbA1c by 1.4% and 56% of participants achieved the ADA goal of glycaemic control, that is, HbA1c < 7.0%.

It should be noted that participants in the present study did not achieve optimal glycaemic control despite a large daily dose of insulin, indicating the presence of severe insulin resistance, consistent with the BMI of 36 kg/m^2. Further, the increased risk of hypoglycaemia associated with escalation of the insulin dose precluded further increase in the daily insulin dose. Thus, the results of the present study demonstrate the multiple metabolic benefits of combining oral agents, for example, pioglitazone and SGLT2i, with insulin therapy in poorly controlled insulin-treated T2DM patients. This strategy can improve glycaemic control and allow significant reduction in daily insulin dose without increased risk of hypoglycaemia or ketosis. It remains to be seen whether the combination of pioglitazone and SGLT2i will exert similar metabolic benefits in insulin-treated lean T2DM patients.

The combination of pioglitazone and dapagliflozin was associated with a low rate of adverse events. The lack of change in body weight can be explained by the glucosuric effect of dapagliflozin, by urinary loss of calories and by the decrease in insulin dose. Only one patient experienced fluid retention which was managed with diuretic therapy. It is probable that the diuretic action of dapagliflozin contributed to the low incidence of fluid retention caused by pioglitazone. However, because of the relatively small number of participants in the present study, a larger study is required to provide a definitive answer to this possibility. Pioglitazone therapy has been associated with a small decrease in haematocrit in some studies.[15] However, in the present study, combination therapy with pioglitazone plus dapagliflozin caused a small (6%) but significant increase in haematocrit. Of note, the increase in haematocrit has been postulated to account for 50% of the cardiovascular benefit of empagliflozin in the EMPA-REG study.[16].

A larger study with a more diverse patient population receiving various background antidiabetic therapies is required to examine the generalizability of the present results to T2DM patients. In summary, the present study demonstrates that combination therapy with pioglitazone plus SGLT2i is a very effective and safe therapeutic strategy in poorly controlled insulin-treated T2DM patients, especially those receiving a high insulin dose.

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CONFLICT OF INTEREST

R. A. D. has served on Advisory Boards for Astra Zeneca, Novo Nordisk, Janssen, Intarcia and Boehringer-Ingelheim; has received research support from Bristol Myers Squibb, Boehringer-Ingelheim, Takeda and Astra Zeneca; has served on Speakers Bureaus for Novo Nordisk and Astra Zeneca. All other authors have no conflict of interest.

Authors contributions

O. M., A. M., M. F. and D. K. generated data for the study. M. A. G designed the study, wrote the protocol, contributed to data generation and data analysis, and wrote the manuscript. R. S. contributed to statistical analysis. R. A. D. and A. J. reviewed and revised the manuscript. M. A. G is the guarantor of this work and, as such, had full access to all the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

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