SGLT2 Inhibition Does Not Affect Myocardial Fatty Acid Oxidation or Uptake, but Reduces Myocardial Glucose Uptake and Blood Flow in Individuals With Type 2 Diabetes: A Randomized Double-Blind, Placebo-Controlled Crossover Trial

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Sodium–glucose cotransporter 2 (SGLT2) inhibition reduces cardiovascular morbidity and mortality in individuals with type 2 diabetes. Beneficial effects have been attributed to increased ketogenesis, reduced cardiac fatty acid oxidation, and diminished cardiac oxygen consumption. We therefore studied whether SGLT2 inhibition altered cardiac oxidative substrate consumption, efficiency, and perfusion. Thirteen individuals with type 2 diabetes were studied after 4 weeks' treatment with empagliflozin and placebo in a randomized, double-blind, placebo-controlled crossover study. Myocardial palmitate and glucose uptake were measured with ^{11}C-palmitate and ^{18}F-fluorodeoxyglucose positron emission tomography (PET)/computed tomography (CT). Oxygen consumption and myocardial external efficiency (MEE) were measured with ^{11}C-acetate PET/CT. Resting and adenosine stress myocardial blood flow (MBF) and myocardial flow reserve (MFR) were measured using ^{15}O-H_2O PET/CT. Empagliflozin did not affect myocardial free fatty acids (FFAs) uptake but reduced myocardial glucose uptake by 57% ($P < 0.001$). Empagliflozin did not change myocardial oxygen consumption or MEE. Empagliflozin reduced resting MBF by 13% ($P < 0.01$), but did not significantly affect stress MBF or MFR. In conclusion, SGLT2 inhibition did not affect myocardial FFA uptake, but channeled myocardial substrate utilization from glucose toward other sources and reduced resting MBF. However, the observed metabolic and hemodynamic changes were modest and most likely contribute only partially to the cardioprotective effect of SGLT2 inhibition.

Treatment with sodium–glucose cotransporter 2 (SGLT2) inhibitors has consistently shown marked reductions in the risk of cardiovascular disease in individuals with type 2 diabetes (1–3). However, SGLT2 inhibitor treatment is also associated with substantial reductions in cardiovascular events in individuals with heart failure without diabetes, where the effect on plasma glucose is minimal (4). The cardioprotective effects of SGLT2 inhibitors can therefore not be mediated merely through lowering of blood glucose levels. However, exactly which alternative mechanisms may be responsible for such beneficial effects of SGLT2 inhibition remain unclear (5).

It has been suggested that SGLT2 inhibitors exert their beneficial effect through an increase in ketogenesis (6). A shift toward myocardial ketone body oxidation potentially improves myocardial energetics (7), because ketone bodies require less oxygen to generate ATP than the quantitatively most important myocardial substrate, free fatty

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acids (FFAs) (7). In addition, increased levels of circulating ketone bodies may also have other potential beneficial effects, because elevated ketone body levels have been demonstrated to increase myocardial perfusion (8), ejection fraction (9), and cardiac output (CO) (9). Another potential cardioprotective mechanism is through the natriuresis and osmotic diuresis induced by SGLT2 inhibitors (10). The diuretic effect reduces extracellular fluid volume and blood pressure leading to a reduction in cardiac preload and afterload (5), which may improve left ventricular function and reduce cardiac workload and oxygen demand (5).

To further explore the cardioprotective effects of SGLT2 inhibitors, we performed a 4-week randomized, double-blind, placebo-controlled crossover trial of empagliflozin (EMPA) to determine the effects on myocardial substrate metabolism, oxygen consumption, and perfusion in individuals with type 2 diabetes. We used a range of positron emission tomography (PET) tracers to measure myocardial FFAs and glucose utilization, myocardial oxygen consumption, myocardial external efficiency (MEE), myocardial blood flow (MBF), and myocardial flow reserve (MFR) in vivo. We hypothesized that SGLT2 inhibition would induce a metabolic shift from FFA and glucose toward oxidation of other substrates, leading to a reduced myocardial FFA oxidation and a reduced myocardial FFA and glucose uptake. We also hypothesized that SGLT2 inhibition would reduce cardiac workload and MBF secondary to a reduction in cardiac afterload.

RESEARCH DESIGN AND METHODS

The study was approved by the Danish Medicines Agency (København S, Denmark) and the regional ethics committee (Viborg, Denmark). The study was registered at eudract .ema.europa.eu (EUDRA-CT nr: 2017-001779-22). Informed, written consent was obtained from all participants.

Trial Design and Participants

The study was a randomized double-blind, placebo-controlled crossover study of once-daily EMPA 25 mg. Each treatment period was 4 weeks with a 1-week washout in between. Inclusion criteria were *1*) type 2 diabetes, *2*) diabetes duration >1 year, *3*) HbA_{1c} 6.5–9.0% (48–75 mmol/mol), *4*) age 50–70, *5*) metformin as only antidiabetic pharmacological treatment, and *6*) unchanged glucose-lowering treatment for 3 months (1 month for other medications). Exclusion criteria were *1*) active or prior cancer, *2*) impaired renal function (estimated glomerular filtration rate <60 mL/min), *3*) recent myocardial infarction (<1 year), *4*) anemia (hemoglobin <6.5 mmol/L), *5*) recurrent genital infections, *6*) prior ketoacidosis, and *7*) alcohol abuse. Participants were recruited through advertisements in local press. All participants underwent a physical examination, provided routine blood samples, and underwent electrocardiography to evaluate eligibility. Participants were randomly assigned in a 1:1 ratio to receive EMPA or placebo in the first study period. Randomization and encapsulation of medicine were handled by the hospital pharmacy. Excess trial medication was returned, and the remaining number of capsules was counted to ensure compliance. Participants' blood samples were examined once weekly.

Three days before the end of each study period, participants were studied after an overnight fast. During this visit, a whole-body DXA (Horizon, Hologic and Discovery; Hologic) scan was performed to assess body composition and indirect calorimetry (Jaeger Oxycon Pro, Intramedic, and Deltatrac II; Datex) to measure resting energy expenditure (EE) and respiratory quotient (RQ). In addition, flash glucose monitoring (Flash Libre; Abbott, Chicago, IL) was applied for the 72 h preceding the PET/CT study day. Measurements from the 1st day were discarded due to the greater imprecision of measurements during the first 24 h. To prevent impact of results on patient behavior, the display of the reader unit was blinded with black tape. Finally, equipment for measuring 24-h blood pressure (Arteriograph 24; Tensiomed, Budapest, Hungary) and activity level (Fitbit Charge 2; Fitbit, San Francisco, CA) were applied. Participants were instructed to maintain an activity level equal to 6,000–10,000 steps/day to ensure comparable activity levels in the two study periods.

PET Protocols and Data Acquisition

Participants were studied in the postabsorptive state after an overnight fast and took the last dose of EMPA/placebo in the morning of the PET/CT study day. PET/CT examinations were done on a Siemens Biograph TruePoint TrueV 64 or a Siemens Biograph Vision (Siemens Healthcare, Erlangen, Germany). Images were reconstructed with a voxel size of $4 \times 4 \times 4$ mm, and all participants underwent PET/CT examinations on the same PET scanner on both study days. Due to tracer production difficulties, not all PET examinations were performed on all participants. The number of successful PET examinations are listed below and in the figure legends for each radiotracer.

Participants were placed with the heart in the field of view, and a low-dose CT scan (16 mA, 100 kV) was obtained for attenuation and anatomic localization purposes. Timing of the PET/CTs was planned to allow for radiotracer decay. The PET/CT flowchart is displayed in Fig. 1*B*, and a detailed description of PET protocols and data acquisition are available in the Supplementary Material.

PET Image Analysis

Dynamic PET image analyses were performed using the in-house developed software package aQuant Research by an assessor blinded to study day sequence. PET image analysis is described in the Supplementary Material.

Blood Samples

Glucose was analyzed immediately after sampling using YSI 2300 STAT Plus glucose analyzer (YSI Life Sciences, Yellow Springs, OH). 3-Hydroxybutyrate (3-OHB) samples were stored at −20°C, and other samples were

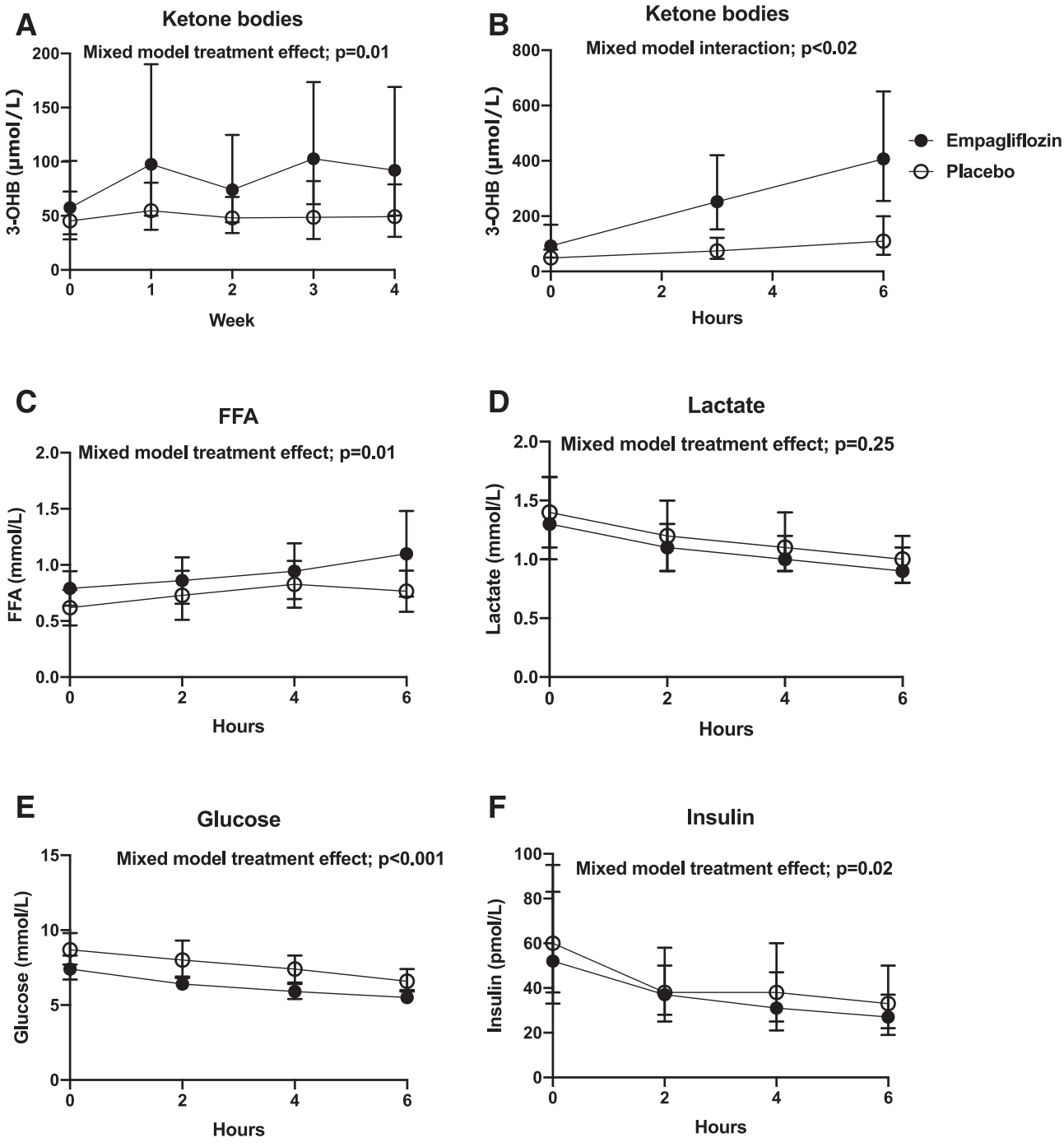

Figure 1—3-OHB concentration during the study period and substrate concentrations during the study day. *A*: EMPA increased 3-OHB concentration from week 1 during the 4-week intervention compared with placebo. Data are plotted as median with 95% CI. *B*: The 3-OHB concentration was higher from the start of the study day and increased more during the study day after EMPA compared with placebo. Data are plotted as medians with 95% CI. *C*: EMPA increased FFA concentration during the study day compared with placebo. Data are mean with 95%. *D*: Lactate decreased during the study day but was similar during EMPA and placebo. Data are median with 95% CI. *E*: Plasma glucose was lower on the study day during EMPA compared with placebo. Data are median with 95%. *F*: Insulin levels decreased during both study days and were lower during EMPA treatment. Data are median with 95% CI. Data were analyzed using linear mixed model analysis.

stored at −80°C until batch analysis. 3-OHB concentrations in serum were quantified using liquid chromatography tandem mass spectrometry (11), serum FFA concentrations with an enzymatic colorimetric method assay nonesterified fatty acids (NEFA)-HR (Wako Chemicals GmbH, Neuss, Germany), lactate concentrations with immobilized enzyme biosensor technology (YSI 2300 model Stat Plus; YSI Life Sciences), and insulin with the AutoDELFIA immunoassay (PerkinElmer, Waltham, MA).

Statistics

Data are presented as mean ± SD or median (95% CI) as appropriate. Data were inspected with QQ-plots for normal distribution, and data were log transformed when appropriate. Paired-samples *t* test was used to detect effects of EMPA compared with placebo on PET/CT measurements. Mixed model analysis was used for repeated measurements during the study period (3-OHB) and during the study days (3-OHB, glucose, FFA, lactate) with treatment, time, and the interaction between treatment and time as fixed factors. *P* values of <0.05 were considered statistically significant. The primary end point was the myocardial FFA oxidation rate since we hypothesized a shift from FFA to ketone body oxidation. To detect a clinically significant reduction of 20% in FFA oxidation, a sample size of 10 subjects was required. To account for potential missing values, we planned to include 12 subjects. The remaining outcomes, except left ventricular mass and ejection fraction, were predefined in our protocol as secondary outcomes.

Data and Resource Availability

The data are available from the corresponding author upon reasonable request. No applicable resources were generated or analyzed.

Table 1—Patient characteristics

	Mean ± SD or *n* (%)
Age (years)	62 ± 6
BMI (kg/m^2)	31.5 ± 5.0
Diabetes duration (years)	4.6 ± 3.0
HbA_{1c} (%)	7.3 ± 2.7
HbA_{1c} (mmol/mol)	56.7 ± 5.5
Sex	
Male	10 (77)
Female	3 (23)
Race	
White	12 (92)
Black or African American	1 (8)
Ischemic heart disease	1 (8)
Antidiabetic drugs	
Metformin	13 (100)
Antihypertensive drugs	
RAAS inhibitor	8 (62)
Thiazide	2 (15)
β-Blocker	2 (15)
Calcium channel blockers	2 (15)
Lipid-lowering drugs	8 (62)

RAAS, renin-angiotensin-aldosterone system.

RESULTS

The study screened 23 volunteers for eligibility and 13 participants were included. The study was planned to include 12 participants, but an extra participant was recruited after 1 participant withdrew consent after the first study day due to claustrophobia during PET scans. Baseline characteristics are presented in Table 1.

Metabolic Variables, Blood Pressure, Body Composition, and Energy Expenditure

EMPA reduced 48-h mean glucose measured by FGM (8.0 ± 0.9 vs. 9.4 ± 2.2 mmol/L, $P < 0.01$) and 24-h systolic (124 ± 8 vs. 129 ± 12 mmHg, $P < 0.05$) and diastolic blood pressure (70 ± 6 vs. 74 ± 7 mmHg, $P < 0.05$) compared with placebo. Body weight was unaltered (94.6 ± 9.6 vs. 95.2 ± 9.7 kg, $P = 0.15$), but EMPA led to a decrease in lean body mass (59.4 ± 5.6 vs. 60.4 ± 5.4 kg, $P = 0.03$). Total body fat mass (31.4 ± 12.2 vs. 31.2 ± 11.3 kg, $P = 0.53$) and fat percentage (32.9 ± 10.1 vs. 32.4 ± 9.3%, $P = 0.26$) were similar after EMPA and placebo. EE was similar (7,435 ± 544 vs. 7,443 ± 481 kJ/day, $P = 0.95$), but RQ decreased during EMPA treatment (0.81 ± 0.03 vs. 0.83 ± 0.03, $P = 0.02$). Plasma creatinine (75 ± 17 vs. 70 ± 17 μmol/L, $P = 0.01$) and hematocrit (0.42 ± 0.02 vs. 0.41 ± 0.03, $P < 0.01$) increased during EMPA.

EMPA treatment increased FFA (0.86 ± 0.30 vs. 0.72 ± 0.27 mmol/L, $P = 0.02$) and 3-OHB concentration (92 (95% CI 50–169) vs. 49 (95% CI 31–79) μmol/L, $P < 0.01$) compared with placebo. The increase in 3-OHB was present from week 1 and remained stable throughout the treatment period (treatment: $P = 0.01$, time: $P = 0.05$, interaction: $P = 0.47$) (Fig. 1*A*). 3-OHB increased more during the study day with EMPA and was 83% (22–173%) higher at the beginning of the study day and 226% (117–390%) higher at the end of the study day (interaction: $P = 0.02$) compared with placebo (Fig. 1*B*). FFA concentration increased during the study day and was consistently higher during EMPA than during placebo (treatment: $P = 0.01$; time effect: $P = 0.01$; interaction: $P = 0.42$) (Fig. 1*C*). Lactate levels were unaffected by EMPA but decreased during the study day (treatment: $P = 0.25$; time: $P < 0.01$; interaction: $P = 0.86$) (Fig. 1*D*). Plasma glucose concentration decreased over the study day and was consistently lower during EMPA (treatment: $P < 0.001$; time effect: $P < 0.0001$; interaction: $P = 0.20$) (Fig. 1*E*). Insulin levels were lower after EMPA (65 ± 47 vs. 84 ± 51 pmol/L, $P = 0.01$) and were consistently lower throughout the study day (treatment: $P = 0.02$, time effect: $P < 0.0001$; interaction: $P = 0.31$) (Fig. 1*F*).

Myocardial ^{11}C-Palmitate Metabolism

EMPA treatment reduced the relative myocardial FFA uptake rate (8.0 ± 1.4 vs. 9.8 ± 1.7 mL/100 g/min, $P < 0.001$) and relative myocardial FFA oxidation rate (6.9 ± 1.2 vs. 8.7 ± 1.7 mL/100 g/min, $P < 0.001$), with no change in relative myocardial FFA reesterification rate (1.2 ± 0.4 vs. 1.2 ± 0.5 mL/100 g/min, $P = 0.98$) (Fig. 2*A*). When absolute metabolism rates were calculated by multiplying the relative metabolism rates by plasma FFA concentrations, similar myocardial FFA uptake rate (7.7 ± 3.7 vs. 8.2 ± 3.6 μmol/100 g/min, $P = 0.75$), myocardial FFA oxidation rate (primary end point) (6.6 ± 3.2 vs.

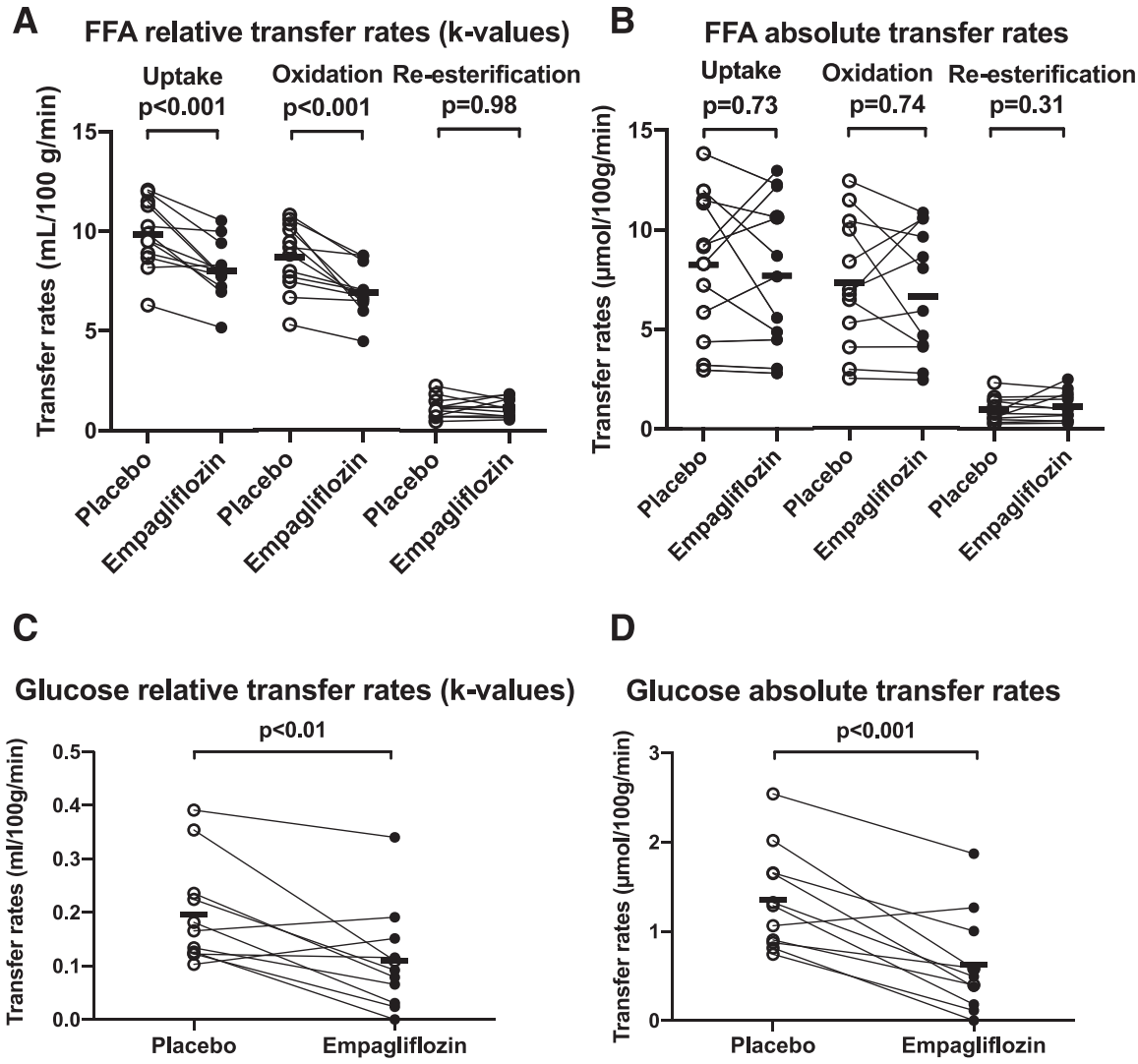

Figure 2—Myocardial FFA and glucose metabolism. *A*: EMPA reduced FFA relative uptake and oxidation rate compared with placebo. *B*: Absolute FFA metabolism rates did not change during EMPA compared with placebo ($n = 12$). EMPA reduced both relative myocardial glucose uptake rate (*C*) and absolute myocardial glucose uptake rate (*D*) compared with placebo ($n = 11$). Data were analyzed with a paired samples *t* test.

7.3 ± 3.3 μmol/100 g/min, $P = 0.54$), and myocardial FFA reesterification rate (1.1 ± 0.7 vs. 1.0 ± 0.6 μmol/100 g/min, $P = 0.31$) were observed (Fig. 2*B*).

Myocardial ^{18}F-Fluorodeoxyglucose Uptake

EMPA treatment reduced relative myocardial glucose uptake rate (0.11 ± 0.10 vs. 0.20 ± 0.10 mL/100 g/min, $P < 0.01$) (Fig. 2*C*). When absolute myocardial glucose uptake was calculated by multiplying relative uptake rates with the plasma glucose concentration, EMPA treatment reduced absolute myocardial glucose uptake by >50% (0.6 ± 0.6 vs. 1.4 ± 0.6 μmol/100 g/min, $P < 0.001$) (Fig. 2*D*).

MBF

EMPA decreased resting MBF (0.74 ± 0.10 vs. 0.85 ± 0.10 mL/g/min, $P < 0.01$), but did not significantly affect stress MBF (3.08 ± 0.79 vs. 3.10 ± 0.80 mL/g/min, $P = 0.92$), MFR (4.14 ± 1.06 vs. 3.60 ± 0.86, $P = 0.09$), or CO (5.6 ± 0.9 vs. 5.5 ± 0.8 L/min, $P = 0.55$) (Fig. 3*A–D*). Since MBF is dependent on cardiac work, we also analyzed the data after adjustment for rate pressure product (pulse × systolic blood pressure/10,000). Here, we also observed a reduction in resting MBF (1.01 vs. 1.06 mL/g/min, $P = 0.04$).

Myocardial Oxygen Consumption, MEE, Left Ventricular Mass, and Ejection Fraction

EMPA did not significantly change myocardial oxygen consumption (8.8 ± 1.0 vs. 9.7 ± 1.4 mL/100 g/min, $P = 0.12$) or MEE (29.5 ± 7.7 vs. 27.0 ± 4.1%, $P = 0.22$) (Fig. 3*E* and *F*).

EMPA did not affect left ventricular mass (127 ± 29 vs. 134 ± 24 g, $P = 0.67$) or ejection fraction (69 ± 7 vs. 70 ± 7%, $P = 0.90$).

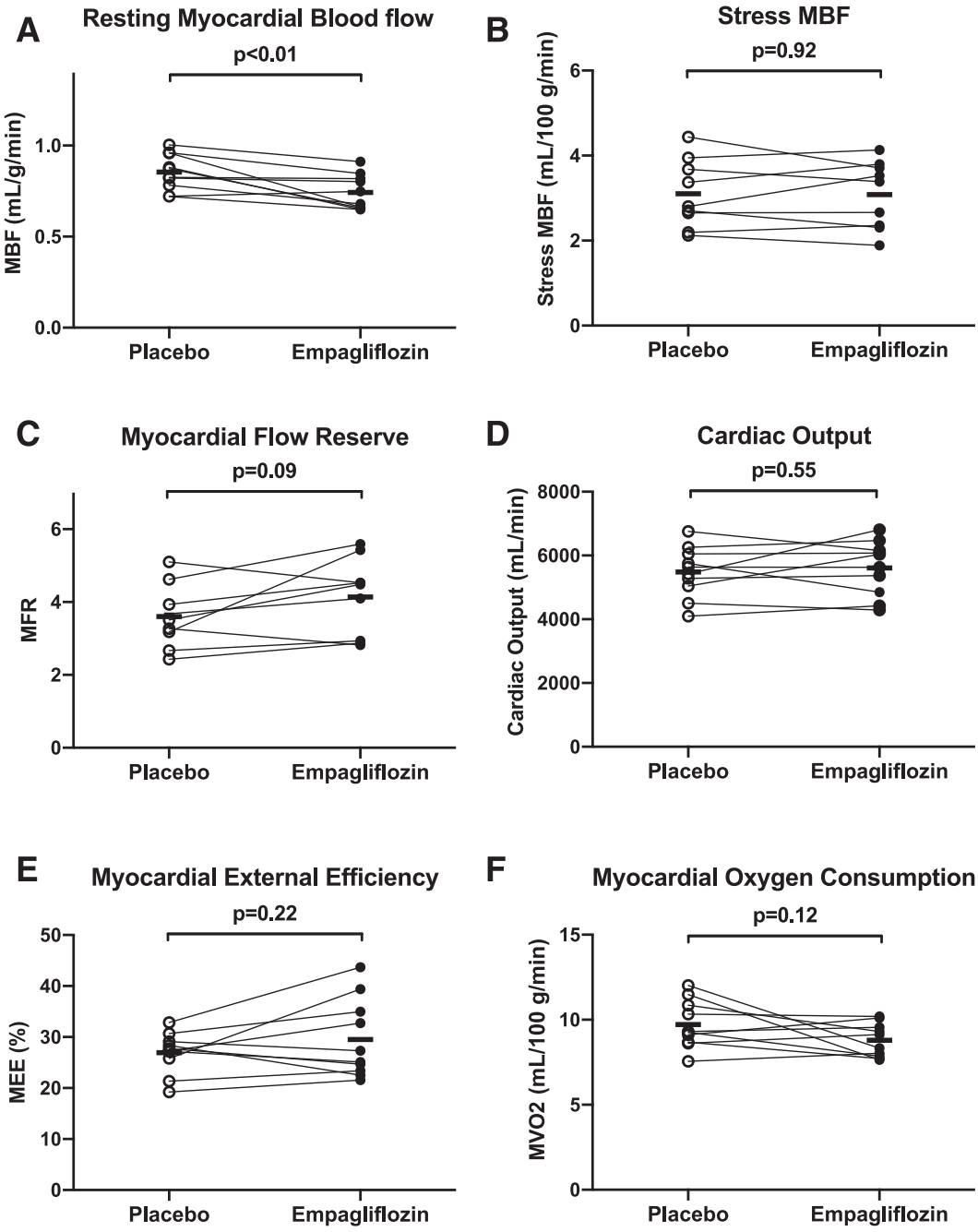

Figure 3—MBF, CO, MEE, and myocardial oxygen consumption. *A*: EMPA reduced resting MBF compared with placebo. EMPA did not significantly affect stress MBF (*B*), MFR (*C*), and CO (*D*) (MBF: $n = 10$, stress MBF: $n = 9$). *E* and *F*: MEE and myocardial oxygen consumption were unaffected by EMPA compared with placebo ($n = 10$). Data were analyzed with a paired samples *t* test.

DISCUSSION

The current study was performed to determine the effects of SGLT2 inhibition on cardiac substrate metabolism, oxygen consumption, and perfusion in individuals with type 2 diabetes. We specifically aimed to investigate whether changes in cardiac intermediary metabolism could be involved in the cardioprotective effects of SGLT2 inhibitors. First, EMPA did not affect myocardial FFA metabolism but reduced the low myocardial glucose uptake by 57%, presumably due to an increase in oxidation of other substrates. Second, the EMPA-driven shift in substrate utilization did not result in measurable changes in myocardial oxygen consumption or MEE. Third, EMPA reduced resting MBF by 13%, which was significant even after adjustment for cardiac workload. Collectively, these results indicate that SGLT2 inhibition is associated with an altered composition of oxidative energy substrates, but these changes are quantitatively of a minor degree and are unlikely to

explain the cardioprotective effect of SGLT2 inhibition. Of interest, SGLT2 inhibition reduced resting myocardial perfusion, but the mechanisms behind this remain to be elucidated.

EMPA Reduces Myocardial Glucose Uptake, but Not FFA Uptake or Oxidation

SGLT2 inhibition increases ketogenesis and concentrations of circulating ketone bodies moderately (6). Recently, it has been hypothesized that such increases in circulating ketone bodies could serve as an oxygen-sparing energy-efficient substrate for the heart, since oxidation of ketone bodies requires less oxygen to generate the same amount of ATP compared with FFA (the thrifty substrate hypothesis) (7). We have previously shown that acute experimental hyperketonemia in healthy subjects results in glucose being replaced by ketone bodies as the preferred myocardial oxidative substrate (8). However, that study was performed during a hyperinsulinemic-euglycemic clamp, with lipolysis and circulating FFAs suppressed to a minimum coupled with maximal myocardial glucose uptake. This particular metabolic milieu prevented us from determining whether ketone bodies could also replace FFA as a substrate for the heart. To address this question, our current study was designed to investigate the effect of SGLT2 inhibition on cardiac FFA metabolism in the physiological, postabsorptive range. Therefore, participants were investigated after an overnight fast to avoid insulin-mediated suppression of ketogenesis and lipolysis. Consequently, we observed 10-fold higher FFA levels than in our previous study. As observed in most other SGLT2 inhibitor studies (6,12), EMPA treatment resulted in significantly increased circulating FFAs compared with placebo, but somewhat surprisingly, absolute rates of myocardial fatty acid utilization, oxidation, and esterification were unaltered. Because circulating FFA concentration is the primary driver of FFA uptake and oxidation (13,14), an increase in myocardial fatty acid utilization and myocardial fatty acid oxidation might have been expected. However, despite similar absolute FFA uptake and oxidation rates, we observed a decrease in relative fatty acid uptake and oxidation capacity (k-values). This downregulation may be mediated through the regulation of fatty acid translocase (FAP)/CD36, the primary transport protein for myocardial FFA uptake (15). FAP/CD36 is regulated by FFA and insulin concentration, and the higher FFA and lower insulin concentration observed during SGLT2 inhibition may have increased FAP/CD36 degradation and thereby reduced myocardial FFA uptake capacity (16).

Whereas EMPA treatment had no effect on fatty acid metabolism, EMPA reduced myocardial glucose uptake >50%. This finding is in accordance with a study in pigs, where SGLT2 inhibition increased ketone body and FFA uptake at the expense of glucose after iatrogenic myocardial infarction (17). Therefore, even though the increased delivery of ketone bodies appears to be readily used in the myocardium, this does not occur at the expense of less oxygen-efficient FFA but as a substitute for glucose. However, the reduction in glucose uptake could also reflect an altered oxidation of other cardiac substrates. Recently, it was shown that myocardial branched chain amino acid uptake was increased by EMPA (17), but because amino acid oxidation only accounts for a very small percentage of cardiac substrate oxidation (18), it is unlikely to explain the findings of this study. Another important cardiac substrate is lactate, but we did not observe any difference in lactate concentrations indicating similar lactate oxidation on the two study days. Finally, increased oxidation of glucose from myocardial glycogenolysis or circulating or intramyocellular triglyceride could have replaced plasma glucose as an oxidative substrate. All in all, we find that the most likely explanation for the reduction in glucose uptake is a shift toward ketone body oxidation due to an increased availability of ketone bodies.

Even though we observed a significant reduction in myocardial glucose uptake during EMPA, the absolute uptake rates were 10-fold lower (1.4 vs. 0.8 μmol/100 g/min) than uptake rates observed in studies of healthy subjects, where fasting myocardial glucose uptake is in the range from 10 to 20 μmol/100 g/min when measured with PET (19) or isotopic tracer infusion (20). It has previously been observed that fasting myocardial glucose uptake is reduced in type 2 diabetes (21), but the magnitude observed in this study was surprising to us. In fact, we found that myocardial glucose uptake only contributes ~2% of the energy required to the total cardiac work (cardiac work ~20 J per mL O_2 [22]) during placebo and ~1% during EMPA. This is lower than reported for healthy individuals, where the myocardial glucose uptake has been estimated to account for ~8% of cardiac work when measured as arteriovenous differences over the heart (23). Therefore, glucose appears to contribute minimally as a cardiac substrate in the postabsorptive state in individuals with type 2 diabetes. Based on these findings, we find it unlikely that the observed reduction in myocardial glucose uptake during EMPA is an important mediator of the cardioprotective effect.

Cardiac Oxygen Consumption or MEE

MEE is the ratio between cardiac workload and oxygen consumption, and a reduction in MEE typically reflects a condition in which oxygen consumption increases more than cardiac work (24). MEE is reduced in heart failure (9) and is considered as part of the pathogenesis for heart failure (25). As discussed above, SGLT2 inhibition could potentially improve MEE through a shift toward less oxygen-demanding substrates. However, only limited data from animal studies are available regarding the effect of SGLT2 inhibition on MEE and cardiac oxygen consumption. Two recent studies have shown that EMPA treatment improves MEE in pigs after myocardial infarction (17) and during acute myocardial ischemia (26). In the latter study, MEE increased independently of oxygen consumption and myocardial fuel switching. This implies that SGLT2

inhibition affects MEE via mechanisms not necessarily mediated through the putative shift in cardiac substrate utilization. SGLT2 inhibition could also affect MEE through the reduction in blood pressure and thus cardiac work, which is supported by data showing that MEE is reduced in patients with hypertension and ventricular hypertrophy (27). In this study, the 4-week intervention with EMPA did not significantly affect cardiac oxygen consumption or MEE. However, although not statistically significant, we did observe a decrease in oxygen consumption ($P = 0.12$) and an increase in MEE ($P = 0.23$), and it is tempting to speculate that a subtle decrease in oxygen consumption of 15% could have been picked up by a study primarily powered to assess ^{11}C-acetate PET (28). Also, we performed ^{11}C-acetate PET as one of the first scans during the study day, where the difference in ketone body concentration was smaller than at the end of the study day. To further complicate our ability to detect an effect on MEE, none of our participants had heart failure as judged by their normal left ventricular ejection fraction and MEE, rendering it unlikely to see an improvement in MEE. It is therefore possible that the results would have been different if subjects with established heart failure and a reduced MEE had been included. It is also possible that the relatively short duration of the intervention partly explains the absence of effect of EMPA on MEE. There are indications that longer-term EMPA treatment results in left ventricular remodeling secondary to reduction in preload and afterload (17,29). However, we observed no change in left ventricular mass, indicating that the intervention period could be too short to detect such morphological changes. In conclusion, additional studies are required to determine whether SGLT2 inhibition improves MEE in individuals with and without heart failure.

EMPA Reduces Resting MBF

Myocardial perfusion is predictive of cardiovascular morbidity and mortality (30). Resting MBF increases with age (31) and primarily depends on cardiac workload and oxygen demand (32). The MFR (ratio between resting and stress MBF) reflects the combined function of epicardial arteries and the myocardial microcirculation, and a reduced MFR is predictive of heart failure (33), cardiovascular events (34), and cardiovascular mortality (30). In the current study, EMPA reduced resting MBF but had no significant effect on stress MBF or MFR, although the latter showed a tendency toward improvement ($P = 0.09$). Improved MFR driven by increased stress-induced hyperemia has previously been demonstrated after 10 weeks of SGLT2 inhibition in prediabetic mice (35), but no studies in humans have thus far been published. Our observation of a reduction in resting MBF during SGLT2 inhibition could very well be caused by the reduced blood pressure. However, resting MBF was still reduced in the EMPA period after adjustment for cardiac workload (rate pressure product), indicating that the decrease in resting MBF is not merely a consequence of reduced cardiac afterload.

In our previous studies, experimental acute hyperketonemia increased resting MBF (8) and CO (9). The increase in CO was dose dependent, with a significant increase at 3-OHB concentrations as low as 0.7 mmol/L (9). By contrast, 4 weeks of EMPA treatment resulted in a more modest increase in 3-OHB to ~0.1–0.2 mmol/L, which had no effect on CO, and resting MBF was actually decreased. These observations appear to suggest that the moderate ketosis induced by SGLT2 inhibition does not have a quantitatively important impact on cardiac hemodynamics.

Strengths and Limitations

A significant strength of this study is the comprehensive in vivo characterization of the effects of EMPA on MBF, oxygen consumption, and substrate metabolism in humans using a range of robust PET/CT tracers and validated kinetic models. However, the study also has limitations. First, the sample size is small, which may have limited our possibility to detect more subtle effects of EMPA on our secondary end points. Second, only one participant had a history of cardiovascular disease. We did not restrict our inclusion criteria to individuals with heart failure or cardiovascular disease to investigate the cardiac effects of SGLT2 inhibitor treatment as second line therapy for the broad population of individuals with type 2 diabetes. Therefore, results could have been different if more participants with cardiovascular disease, especially heart failure, had been included. Subgroup analyses of the Dapagliflozin Effect on Cardiovascular Events trial (DECLARE-TIMI 58) (36) indicated that the beneficial effect of SGLT2 inhibition was primarily observed in subjects with established cardiovascular disease. However, a meta-analysis has shown that the cardiovascular benefit of SGLT2 inhibitors appear to be independent of a history of heart failure (37). Third, we performed an adenosine stress test in the beginning of the study day, which potentially could have affected the estimates from the subsequent PET scans. However, because our two study days were identical and the participants served as their own controls, this is unlikely to explain the observed effects of EMPA.

Conclusion

SGLT2 inhibition does not affect myocardial FFA oxidation or uptake, but induces a shift in myocardial substrate utilization from glucose toward other sources, possibly ketone bodies. However, this shift in myocardial substrate utilization is quantitatively of minor importance and does not appear to improve either MEE or myocardial oxygen consumption. Therefore, it is unlikely to explain the striking cardioprotective benefit of SGLT2 inhibition. Of interest, SGLT2 inhibition reduces resting MBF, even when adjusting for cardiac workload.

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