Diabetologia (2021) 64:42–55
https://doi.org/10.1007/s00125-020-05306-1

ARTICLE

The effects of dapagliflozin, metformin or exercise on glycaemic variability in overweight or obese individuals with prediabetes (the PRE-D Trial): a multi-arm, randomised, controlled trial

Kristine Færch[1] · Martin B. Blond[1] · Lea Bruhn[1] · Hanan Amadid[1] · Dorte Vistisen[1] · Kim K. B. Clemmensen[1] · Camilla T. R. Vainø[1] · Camilla Pedersen[1] · Maria Tvermosegaard[1,2] · Thomas F. Dejgaard[1] · Kristian Karstoft[3] · Mathias Ried-Larsen[3,4] · Frederik Persson[1] · Marit E. Jørgensen[1,2]

Received: 22 May 2020 / Accepted: 28 August 2020 / Published online: 16 October 2020

Abstract

Aims/hypothesis We aimed to investigate the short-term efficacy and safety of three glucose-lowering interventions in overweight or obese individuals with prediabetes defined by HbA_{1c}.

Methods The PRE-D Trial was a randomised, controlled, parallel, multi-arm, open-label, non-blinded trial performed at Steno Diabetes Center Copenhagen, Gentofte, Denmark. One hundred and twenty participants with BMI ≥25 kg/m^2, 30–70 years of age, and prediabetes (HbA_{1c} 39–47 mmol/mol [5.7–6.4%]) were randomised 1:1:1:1 to dapagliflozin (10 mg once daily), metformin (1700 mg daily), interval-based exercise (5 days/week, 30 min/session) or control (habitual lifestyle). Participants were examined at baseline and at 6, 13 and 26 weeks after randomisation. The primary outcome was the 13 week change in glycaemic variability (calculated as mean amplitude of glycaemic excursions [MAGE]) determined using a continuous glucose monitoring system (pre-specified minimal clinically important difference in MAGE ~30%).

Results One hundred and twelve participants attended the examination at 13 weeks and 111 attended the follow-up visit at 26 weeks. Compared with the control group, there was a small decrease in MAGE in the dapagliflozin group (17.1% [95% CI 0.7, 30.8], $p = 0.042$) and a small, non-significant, reduction in the exercise group (15.3% [95% CI −1.2, 29.1], $p = 0.067$), whereas MAGE was unchanged in the metformin group (0.1% [95% CI −16.1, 19.4], $p = 0.991$)). Compared with the metformin group, MAGE was 17.2% (95% CI 0.8, 30.9; $p = 0.041$) lower in the dapagliflozin group and 15.4% (95% CI −1.1, 29.1; $p = 0.065$) lower in the exercise group after 13 weeks, with no difference between exercise and dapagliflozin (2.2% [95% CI −14.8, 22.5], $p = 0.815$). One serious adverse event occurred in the control group (lung cancer).

Conclusions/interpretation Treatment with dapagliflozin and interval-based exercise lead to similar but small improvements in glycaemic variability compared with control and metformin therapy. The clinical importance of these findings in prediabetes is uncertain.

Trial registration ClinicalTrials.gov NCT02695810

Funding The study was funded by the Novo Nordisk Foundation, AstraZeneca AB, the Danish Innovation Foundation, the University of Copenhagen and Ascensia Diabetes Care Denmark ApS

Keywords Dapagliflozin · Exercise · Glycaemic variability · Intermediate hyperglycaemia · Metformin · Overweight · Prediabetes · SGLT2 inhibitor

Electronic supplementary material The online version of this article (https://doi.org/10.1007/s00125-020-05306-1) contains peer-reviewed but unedited supplementary material, which is available to authorized users.

✉ Kristine Færch
kristine.faerch@regionh.dk

[1] Steno Diabetes Center Copenhagen, Gentofte, Denmark

[2] University of Southern Denmark, Copenhagen, Denmark

[3] The Centre of Inflammation and Metabolism and the Centre for Physical Activity Research, Rigshospitalet, University of Copenhagen, Copenhagen, Denmark

[4] The Danish Diabetes Academy, Odense, Denmark

Research in context

What is already known about this subject?

- HbA_{1c} is now commonly used for the diagnosis of prediabetes but owing to poor overlap with former diagnostic criteria results from previous prevention studies may not apply to those identified by this new criterion
- Sodium–glucose cotransporter 2 (SGLT2) inhibitors have proven efficacious in type 2 diabetes but their ability to improve blood glucose levels and cardiovascular risk in comparison with other glucose-lowering interventions has not previously been examined in individuals with prediabetes

What is the key question?

- Do metformin, exercise and SGLT2 inhibitors affect glycaemic variability and other cardiometabolic outcomes in people with HbA_{1c}-defined prediabetes?

What are the new findings?

- Dapagliflozin and interval-based exercise showed similar small effects on glycaemic variability in prediabetes
- Interventions with dapagliflozin, metformin or interval-based exercise were all associated with small improvements in HbA_{1c} and other markers of glucose metabolism
- Adverse events were reported by a majority of those receiving a pharmacological treatment, whereas the rate of adverse events in the exercise group was comparable to that of the control group

How might this impact on clinical practice in the foreseeable future?

- Our results do not support the idea that the benefits of short-term pharmacological interventions with metformin or SGLT2 inhibitors outweigh the harms in overweight or obese individuals with HbA_{1c} levels in the prediabetic range (as defined by the ADA); interval-based exercise is the preferable intervention in this population because of the additional cardiometabolic benefits and few adverse events

Abbreviations

CGM	Continuous glucose monitoring
CRF	Cardiorespiratory fitness
CV_{CGM}	CV of sensor glucose
$iAUC_{glucose}$	Incremental AUC for glucose
$iAUC_{insulin}$	Incremental AUC for insulin
MAGE	Mean amplitude of glycaemic excursions
SD_{CGM}	SD of sensor glucose
SGLT2	Sodium–glucose cotransporter 2

Introduction

Prediabetes is a common metabolic condition aetiologically related to obesity [1] and associated with increased risk of type 2 diabetes, CVD and all-cause mortality [2]. Identifying effective strategies to reduce diabetes risk and improve cardiometabolic function in this population is therefore important. Randomised controlled trials have shown that weight loss, lifestyle modification and metformin treatment can reduce the progression to diabetes in individuals with impaired glucose tolerance [3–5]. However, evidence is lacking in individuals with impaired fasting glucose or prediabetes assessed by HbA_{1c} [6, 7]. Despite this, the ADA suggests that all people with prediabetes should accomplish and maintain weight loss, increase physical activity and consider metformin treatment [8]. HbA_{1c} is now commonly used for the diagnosis of diabetes and prediabetes, but due to the poor overlap between fasting glucose, 2 h glucose and HbA_{1c} [9–12], results from intervention studies in individuals with impaired glucose tolerance may not apply to those with prediabetes diagnosed by HbA_{1c}. To address this question, trials focusing on improving blood glucose levels and reducing cardiovascular risk in people with elevated HbA_{1c} levels are needed.

In recent years, sodium–glucose cotransporter 2 (SGLT2) inhibitors have proven efficacious in reducing blood glucose, body weight and cardiovascular events in individuals with type 2 diabetes [3, 13]. A few smaller studies have examined the effect of SGLT2 inhibition in prediabetes [14–16], suggesting that these drugs have potential in the prevention of type 2 diabetes. However, the ability of SGLT2 inhibitors to improve overall blood glucose levels, glycaemic variability and other cardiometabolic risk factors in individuals with prediabetes in comparison with other glucose-lowering interventions has not previously been examined.

We conducted a randomised controlled trial comparing the short-term safety and efficacy of three glucose-lowering interventions (dapagliflozin, metformin and exercise) in combination with dietary advice on glycaemic variability, body composition and cardiometabolic risk in overweight or obese individuals with prediabetes defined by HbA_{1c} [17]. The

primary outcome was changes in glycaemic variability from baseline to 13 weeks of intervention. Glycaemic variability was assessed by continuous glucose monitoring (CGM) to reflect glucose fluctuations in daily life. Glycaemic variability is an important aspect of glycaemic control, which is associated with morbidity and mortality in type 2 diabetes [18, 19]; however, whether this extends to people with prediabetes is yet unknown. Exercise has previously been shown to reduce glycaemic variability in individuals with type 2 diabetes or who are at increased risk for developing the disease [20, 21]. We hypothesised that after 13 weeks of treatment, exercise would be superior to the pharmacological interventions and dapagliflozin would be superior to metformin in reducing glycaemic variability.

Methods

Trial design

The PRE-D Trial was an investigator-initiated, randomised, controlled, open-label, four-arm (1:1:1:1), superiority trial performed at Steno Diabetes Center Copenhagen, Gentofte, Denmark. The trial was initiated on 24 February 2016 and terminated on 13 January 2019. The design of the PRE-D Trial has previously been published with open access [22]. The trial protocol was approved by the Ethics Committee of the Capital Region (H-15011398) and the Danish Medicines Agency (EudraCT: 2015-001552-30) and registered at ClinicalTrials.gov (NCT02695810). Approval of data storage was obtained from the Danish Data Protection Board (2012-58-0004). The trial was conducted in accordance with the Helsinki II declaration and Good Clinical Practice.

Because of a slow recruitment rate, the inclusion criterion for HbA_{1c} was expanded from 42–47 mmol/mol (6.0%–6.4%) (criterion suggested by the International Expert Committee) to 39–47 mmol/mol (5.7%–6.4%) (criterion suggested by the ADA), effective from 13 April 2016. Also, the number of participants to be included was reduced from 160 to 120 on 11 July 2016.

The trial consisted of a baseline test period (baseline visit and CGM measurement during the subsequent 6 days), 13 weeks of active intervention, and a follow-up period of 13 weeks with no active treatment. Test days and subsequent 6 day CGM measurement periods were scheduled at baseline and at 6, 13 and 26 weeks after intervention start. The trial was designed to examine effects during active treatment (visit at 6 weeks and CGM measurement during the subsequent 6 days) and immediately after the termination of treatment (visit at 13 weeks and CGM measurement during the subsequent 6 days with the primary endpoint measured here). Additionally, potential effects sustained after the end of treatment were examined (follow-up visit at 26 weeks and CGM measurement during the subsequent 6 days).

Participant and public involvement

No participants were involved in defining the research question or the outcome measures, nor were they involved in the implementation of the study. We disseminated the results from the trial to the participants and invited them to participate in focus-group interviews after the end of the study.

Participants

The study included participants aged 30–70 years with overweight or obesity (BMI ≥25 kg/m^2) and who had prediabetes based on the HbA_{1c} criterion (39–47 mmol/mol [5.7–6.4%]) [23]. Exclusion criteria were as follows: pregnancy (planned or current); breast feeding; known diabetes; bariatric surgery within the past 2 years; neurogenic bladder disorder; impaired renal function (eGFR <60 ml min^{-1} [1.73 m]$^{-2}$); uncontrolled medical issues; use of beta blockers, steroids, loop diuretics, thiazide diuretics or medication affecting glucose metabolism (though treatment with statins was allowed); allergy to one of the medications used in the trial; or being unable to exercise according to the protocol. Participants with hypertension who were taking any of the above-mentioned BP-lowering medications and who were willing to switch to a drug allowed in the trial were enrolled.

Participants were recruited using public advertisements and at the Steno Diabetes Center Copenhagen outpatient clinic (relatives and friends to patients with diabetes). All participants provided oral and written informed consent before taking part in the study. Participants received no incentives, monetary or otherwise, for their participation in the PRE-D Trial, although documented travel costs from the participants' homes to the clinic were reimbursed.

Interventions

All participants received general advice on diet and physical activity based on national recommendations [24]. After the baseline test period, participants were randomly assigned to receive 13 weeks of intervention as follows: (1) dapagliflozin, 10 mg once daily; (2) metformin, 850 mg daily for 1 week and then 850 mg twice daily for the rest of the intervention; (3) exercise; or (4) control (habitual living). The exercise intervention consisted of unsupervised interval training, 5 days/week, 30 min/session, with alternating 3 min intervals aiming for intensities of ≥75% and ≤60% of peak heart rate by the end of each interval. To support adherence to the exercise protocol, the participants in the exercise group recorded exercise sessions using a heart rate monitor (Polar V800; Polar Electro, Kempele, Finland). Data from exercise sessions (heart rate, duration, frequency) were uploaded by the participants to an online platform (https://flow.polar.com) and brief feedback

was provided weekly by research staff to the participants based on an evaluation of the individual exercise sessions.

For those allocated to active treatments, the treatment was initiated the day after the end of the baseline CGM measurement period. All active treatments were terminated just before the 13 week visit; participants in the exercise group returned the heart rate monitor at 13 weeks.

Procedures and outcomes

Outcomes The primary efficacy outcome was the change in the mean amplitude of glycaemic excursions (MAGE) [25], a measure of glycaemic variability, from baseline to end of treatment (13 weeks). Secondary outcomes included changes from baseline to mid-point of treatment (6 weeks), end of treatment (13 weeks) and follow-up (26 weeks) for the following variables: MAGE (6 and 26 weeks); mean CGM glucose; SD of sensor glucose (SD_{CGM}); CV of sensor glucose (CV_{CGM}); daily time spent at glucose concentrations >6.1 mmol/l, >7.0 mmol/l, >7.8 mmol/l and >11.1 mmol/l measured by CGM; HbA_{1c}; fasting plasma glucose concentration; fasting serum insulin concentration; body weight; WHR; BP; resting heart rate; and blood lipids. Additional secondary outcomes measured only at baseline, at 13 weeks and at 26 weeks included the following variables: plasma glucose and serum insulin levels during an OGTT; measures of body composition; peak heart rate and cardiorespiratory fitness (CRF). At baseline and after 13 and 26 weeks, a resting ECG was performed prior to exercise testing for safety assessment.

Clinic visits and free-living measurement periods To assess eligibility for inclusion, potential participants attended a screening visit consisting of an interview and a medical examination [22]. On subsequent test days (6, 13 and 26 weeks), the participants arrived at the clinic at 08:00–09:00 hours after an overnight fast of at least 8 h. Participants were asked to abstain from strenuous physical activity for 48 h prior to the examinations at 0, 13 and 26 weeks, and those who were allocated to treatment with either dapagliflozin or metformin were asked not to take the study medication on the morning of the test day at week 13 [22]. The test day visits lasted approximately 5 h and were identical except for the test day at 6 weeks, which was of a shorter duration and did not include the ECG, OGTT and the measurement of CRF and body composition. When participants arrived at the test facility, the length of fasting and the time of their last exercise bout were registered, followed by measurements of body weight, plus waist and hip circumference, as described in detail previously [22]. Then the participants were fitted with a blinded CGM system (iPro2 CGM; Medtronic Denmark, Denmark) for the assessment of MAGE and other measures of glycaemic control in a free-living situation; glucose levels were monitored for six consecutive days with the iPro2 placed on the lower abdomen. To calibrate the CGM systems, the participants measured blood glucose levels using a glucose meter (Contour XT; Ascensia Diabetes Care Denmark, Denmark) at 1 h and 2 h after CGM insertion as well as at home before breakfast, before lunch, before dinner and before bedtime during the CGM measurement periods. MAGE was calculated by taking the arithmetic mean of the blood glucose increases or decreases when both ascending and descending segments exceeded the value of 1 SD of the blood glucose during a 24 h measurement period [25] (see electronic supplementary material [ESM] Methods and ESM Fig. 1 for details).

Next, BP and resting heart rate were measured and a resting ECG was recorded; the latter was used to screen for contraindications to perform the fitness test [22]. Fasting blood samples and urine samples were collected. This was followed by a total body dual-energy x-ray absorptiometry scan (Discovery DXA System; Hologic, MA, USA) to assess body composition. Participants wore light clothing and lay still in a supine position during the scan. A standard 75 g OGTT was then performed with blood sampled from an antecubital vein just before the ingestion of glucose and at 30, 60 and 120 min after ingestion. Participants were instructed to either sit or lie down during the OGTT but could read and listen to music/podcast/audiobooks. In addition, they answered electronic questionnaires administered by the staff. The incremental AUCs for glucose ($iAUC_{glucose}$) and insulin ($iAUC_{insulin}$) were calculated based on the trapezoidal rule as measures of the overall glucose tolerance and insulin response during the OGTT. Finally, CRF ($\dot{V}O_{2peak}$) and peak heart rate were evaluated using an incremental test on a cycle ergometer (Monark LC4, Monark, Sweden) with indirect calorimetry. After a 6 min warm up (3 min at 30 W followed by 3 min at 60 W for women; 3 min at 40 W followed by 3 min at 80 W for men) the workload was increased by 20 W/min for women and 25 W/min for men until exhaustion. From trial initiation until 11 January 2018, a JAEGER Oxycon Pro analyser (Erich JAEGER, Germany) was used to measure oxygen and carbon dioxide exchange (210 tests). Due to an equipment malfunction, a Vyntus CPX (Vyaire, Germany) was used for the remainder of the trial (129 tests). Before leaving the facility, the participants were served a well-deserved lunch and were instructed to measure home blood glucose and fill in the dietary diary for the following days.

Sample analysis Samples for the analysis of plasma glucose concentrations were placed on ice immediately following sampling. All samples were centrifuged shortly after collection at 1610 *g* for 15 min (Sigma 4 K15; Sigma, Germany), except for samples used for analysis of HbA_{1c} and serum insulin concentrations. Samples for serum insulin analysis were centrifuged 30 min after collection. The samples were stored in a refrigerator for the remainder of the test day. Urine

samples were cooled after collection. At the end of each test day, all samples from that day were analysed in-house at Steno Diabetes Center Copenhagen. Serum insulin was analysed using electro-chemiluminescence immunoassay (Cobas e411; Roche Diagnostics, Switzerland). HbA_{1c} was measured by HPLC (Tosoh G8; Tosoh Corporation, Japan). Urinary glucose, plasma glucose, total cholesterol, HDL-cholesterol and triacylglycerols were analysed by colorimetric analysis (Vitros 5600; Ortho Clinical Diagnostics, USA). Plasma VLDL-cholesterol was calculated as plasma triacylglycerols (mmol/l)/2.2, and plasma LDL-cholesterol was calculated based on the Friedewald equation [26]. eGFR was calculated using the CKD-EPI formula [27].

Adverse events and treatment adherence monitoring

Adverse events were systematically registered and rated for severity at the 6, 13 and 26 week visits. For adverse events occurring between visits, the participants were instructed to contact the investigators who registered and rated the severity of the episode.

Adherence to treatment in the metformin and dapagliflozin groups was calculated from the amount of medication returned by the participants at the 6 and 13 week visits. Participants taking ≥80% of their medication were regarded as compliant. Adherence to the exercise intervention was calculated from the number and duration of the exercise sessions performed. Participants fulfilling the compound goal of completing ≥80% of the prescribed exercise sessions and ≥80% of the prescribed exercise volume were designated as adherent.

Sample size

The clinical target for glycaemic variability in prediabetes is unknown [28]. We designed the trial to have 80% power (α 0.05) to detect a minimal clinically important difference of 0.5 mmol/l change in glycaemic variability (MAGE) between two groups from baseline to 13 weeks with an SD of 0.6 mmol/l [22]. In our study population, this corresponds to ~30% difference. Based on this assumption, at least 23 participants in each group were needed. To account for drop-outs, we included 30 participants in each group.

Randomisation

Randomisation was performed in blocks of 16 (4:4:4:4) without any stratification for baseline variables using a web-based system (EasyTrial). When using EasyTrial, the allocation of each participant is not determined until the individual participant is ready for randomisation; accordingly, no randomisation list was generated before the trial started. Participants were randomised at the end of the baseline visit but were blinded to group allocation until the end of the 6 days pre-intervention CGM measurement period. Documents and equipment/medication to be used during the intervention were supplied to the participants in non-see-through bags locked with a coded padlock. The participants received the code when finishing the baseline CGM measurement period and later returned the bags and locks in order to confirm that these had not been forced open.

Statistical methods

We performed pre-specified intention-to-treat analyses including all available data. It was a priori planned to compare the intervention effects between all four groups (six tests) for the primary outcome, MAGE. All pre-specified secondary outcomes were only compared with the control group. A per protocol analysis was conducted for MAGE, including participants attending the 13 week visit who were designated adherent. Outcomes were modelled by linear mixed-effects models with a participant-specific random intercept. The baseline level of the outcome was retained as an outcome in the model. Treatment groups and visits and their interaction were included as fixed effects. Additionally, for the primary outcome a post hoc sensitivity analysis including baseline as a covariate rather than as an outcome was performed (ESM Methods). Assumptions of normality and homogeneity of variances for residuals were assessed with graphical methods. Results for MAGE are presented as estimated mean differences in change with 95% CI and two-sided p values. Secondary outcomes are presented as estimated mean differences in change with 95% CIs; the widths of the CIs have not been adjusted for multiple testing. Outcomes for which the distribution of residuals did not conform to the parametric assumptions of the models were transformed using the natural logarithm ($\log_e$) prior to analyses and results are presented as the ratio between the relative change from baseline to follow-up within the two groups being compared (i.e. if group A changed 20% between visits 1 and 2 [A2/A1 = 1.2] and group B changed 10% [B2/B1 = 1.1], then the relative change between group A and B is 1.2/1.1 = 9.1%). Analyses of body composition, CRF and HDL-cholesterol were adjusted for sex. The analysis of CRF was further adjusted for the type of calorimeter used (ESM Methods, ESM Results and ESM Tables 1–3). All statistical analyses were conducted blinded to group allocation and interpretation of results were performed before unblinding. Statistical analyses were conducted in R version 3.6.0 (The R Foundation for Statistical Computing, www.R-project.org) and SAS version 9.4 (SAS Institute, Cary, NC, USA).

Results

Out of 404 individuals screened, 120 participants were randomly assigned to dapagliflozin (n = 30), metformin (n = 30), exercise (n = 30) or control (n = 30) (Fig. 1). Across groups, 44% were men, the median (Q1, Q3) MAGE was 1.6 (1.4, 2.2) mmol/l, the mean±SD HbA_{1c} was 40.9 ±

Table 1 Demographic and baseline characteristics

Characteristic	Dapagliflozin ($n = 30$)	Metformin ($n = 30$)	Exercise ($n = 30$)	Control ($n = 30$)
Age, years	61.4 ± 8.5	56.7 ± 8.4	57.8 ± 9.9	57.2 ± 9.9
Men, *n* (%)	13 (43)	13 (43)	15 (50)	12 (40)
Self-reported ethnic origin, *n* (%)				
White	29 (97)	29 (97)	27 (90)	27 (90)
Other	1 (3)	1 (3)	3 (10)	3 (10)
Current smoker, *n* (%)	2 (7)	2 (7)	7 (23)	2 (7)
Higher education, *n* (%)	15 (50)	15 (50)	11 (37)	11 (37)
Living with partner, *n* (%)	19 (63)	24 (80)	23 (77)	19 (63)
Family history of diabetes, *n* (%)	16 (53)	16 (53)	15 (50)	17 (57)
Family history of CVD, *n* (%)	18 (60)	18 (60)	18 (60)	16 (53)
Antihypertensive medication, *n* (%)	10 (33)	5 (17)	11 (36)	6 (20)
Lipid-lowering medication, *n* (%)	11 (37)	7 (24)	4 (13)	6 (20)
Antiplatelet agents/DOACs, *n* (%)	2 (7)	2 (7)	1 (3)	1 (3)
Systolic BP, mmHg	135 ± 14	131 ± 17	133 ± 17	135 ± 17
Diastolic BP, mmHg	85 ± 7	84 ± 9	87 ± 9	87 ± 8
Body weight, kg				
Men	103.7 ± 17.6	103.1 ± 16.5	103.9 ± 15.0	99.0 ± 15.1
Women	82.5 ± 13.7	83.0 ± 9.8	86.6 ± 7.7	92.1 ± 25.3
BMI, kg/m^2				
Men	31.6 ± 3.8	31.2 ± 4.5	33.2 ± 4.3	30.4 ± 3.2
Women	30.4 ± 5.1	29.3 ± 3.7	31.8 ± 3.4	34.0 ± 8.4
Waist circumference, cm				
Men	110.5 ± 12.2	109.5 ± 11.8	112.4 ± 8.9	107.5 ± 9.0
Women	98.6 ± 15.4	97.4 ± 11.8	101.7 ± 8.2	104.6 ± 17.2
Body fat, %				
Men	31.6 ± 5.0	30.5 ± 4.0	32.1 ± 4.3	31.5 ± 5.1
Women	43.6 ± 4.0	41.8 ± 4.9	42.8 ± 3.1	44.3 ± 6.1
CRF, ml O_2 min^{-1}; kg^{-1})				
Men	25.2 ± 4.9	28.8 ± 7.4	24.2 ± 4.8	30.0 ± 6.6
Women	21.5 ± 3.4	23.7 ± 5.0	21.9 ± 3.6	20.5 ± 5.4
eGFR, ml min^{-1} [1.73 m]$^{-2}$	84 ± 8	86 ± 7	86 ± 7	85 ± 7
Lipids, mmol/l				
Total cholesterol	5.1 ± 1.1	5.4 ± 1.1	5.3 ± 1.1	4.8 ± 0.9
LDL-cholesterol	3.0 ± 1.0	3.2 ± 1.0	3.2 ± 1.0	2.9 ± 0.9
HDL-cholesterol	1.4 ± 0.4	1.5 ± 0.6	1.2 ± 0.3	1.3 ± 0.4
Triacylglycerols	1.2 (0.9–1.6)	1.2 (0.9–1.8)	1.7 (1.1–2.4)	1.1 (0.8–1.6)
HbA_{1c}				
mmol/mol	41.2 ± 2.0	40.5 ± 2.2	41.1 ± 2.2	40.9 ± 2.8
%	5.9 ± 0.19	5.9 ± 0.20	5.9 ± 0.20	5.9 ± 0.26
Fasting plasma glucose, mmol/l	5.6 ± 0.45	5.6 ± 0.76	5.7 ± 0.62	5.5 ± 0.42
Fasting serum insulin, pmol/l	61 (46–96)	61 (45–85)	87 (67–116)	70 (43–103)
OGTT				
30 min plasma glucose, mmol/l	8.6 ± 1.3	8.2 ± 1.2	9.3 ± 1.5	8.9 ± 1.5
60 min plasma glucose, mmol/l	10.0 ± 2.2	8.6 ± 2.0	10.8 ± 2.0	9.6 ± 2.2
120 min plasma glucose, mmol/l	8.0 ± 1.8	7.0 ± 1.9	7.7 ± 2.3	7.0 ± 2.1
$iAUC_{glucose}$, mmol/l × min	361 ± 153	257 ± 123	403 ± 123	332 ± 154
$iAUC_{insulin}$, pmol/l × min× 10^{-3}	47 (28–61)	43 (32–58)	65 (43–115)	49 (29–83)

Table 1 (continued)

Characteristic	Dapagliflozin ($n = 30$)	Metformin ($n = 30$)	Exercise ($n = 30$)	Control ($n = 30$)
Glycaemic status				
HbA_{1c} 39–41 mmol/mol[a], *n* (%)	17 (57)	22 (73)	15 (50)	19 (63)
HbA_{1c} 42–47 mmol/mol[b], *n* (%)	13 (43)	8 (27)	15 (50)	11 (37)
NFG + NGT	9 (30)	17 (57)	8 (27)	13 (43)
Isolated IFG	6 (20)	5 (17)	9 (30)	10 (33)
Isolated IGT	6 (20)	1 (3)	1 (3)	3 (10)
Combined IFG and IGT	8 (27)	5 (17)	8 (27)	2 (7)
Plasma glucose in diabetic range[c]	1 (3)	2 (7)	4 (13)	2 (7)
MAGE, mmol/l	1.7 (1.5–2.4)	1.6 (1.1–1.8)	1.9 (1.4–2.8)	1.5 (1.2–1.8)
Mean $glucose_{CGM}$, mmol/l	6.3 ± 0.50	6.0 ± 0.48	6.3 ± 0.40	6.2 ± 0.38
SD_{CGM}, mmol/l	0.8 (0.7–1.0)	0.8 (0.6–1.0)	0.9 (0.7–1.2)	0.7 (0.6–0.9)
CV_{CGM}, %	14 (12–17)	13 (10–15)	14 (11–18)	12 (10–15)
Percentage of time spent at >6.1 mmol/l	43.2 (31.7–65.7)	43.8 (19.4–52.4)	52.5 (37.5–62.3)	43.3 (29.2–60.9)
Percentage of time spent at >7.0 mmol/l	15.5 (7.0–25.2)	9.6 (3.0–19.8)	16.9 (7.8–28.0)	8.7 (5.4–22.7)
Percentage of time spent at >7.8 mmol/l	5.3 (1.8–11.2)	2.3 (0.5–6.9)	4.7 (2.4–13.8)	2.4 (1.1–8.2)
Percentage of time spent at >11.0 mmol/l	0.0 (0.0–0.0)	0.0 (0.0–0.0)	0.0 (0.0–0.0)	0.0 (0.0–0.0)

Data are expressed as mean±SD, median (25th–75th percentile) or *n* (%)

[a] HbA_{1c} 5.7–5.9%

[b] HbA_{1c} 6.0–6.4%

[c] Defined by fasting plasma glucose ≥7.0 mmol/l or 120 min plasma glucose ≥11.1 mmol/l

DOAC, direct oral anticoagulant; IFG, impaired fasting glucose (fasting plasma glucose ≥5.6 mmol/l); IGT, impaired glucose tolerance (2 h plasma glucose ≥7.8 mmol/l) NFG, normal fasting glucose; NGT, normal glucose tolerance (2 h glucose)

2.3 mmol/mol (5.9 ± 0.2%), mean±SD BMI was 31.5 ± 5.1 kg/m^2 and mean±SD age was 60 ± 9 years. Baseline characteristics of the participants by randomisation group are shown in Table 1.

The overall retention rate was 93%. A total of 26 participants in the dapagliflozin group and 25 in the metformin group took ≥80% of the administered medication (for further details on adherence to the interventions see ESM Table 4). In the exercise group, 24 individuals performed ≥80% of the required exercise training (ESM Table 5). Median (Q1, Q3) urinary glucose concentration was 122.1 (77.6, 163.7) mmol/l after 6 weeks and 76.4 (20.8, 123.3) mmol/l after 13 weeks of treatment with dapagliflozin, whereas it was <1.1 (<1.1, <1.1) mmol/l at baseline and at 26 weeks in the dapagliflozin group and in the other groups at all time points. Only a few changes in non-trial medicines were reported (ESM Results).

Primary outcome

Compared with the control group, MAGE decreased by 17.1% in the dapagliflozin group from baseline to 13 weeks; the effect in the exercise group was smaller and more uncertain (Fig. 2, Table 2). MAGE did not change in the metformin group compared with control. Results from the per protocol analysis and the post hoc baseline-adjusted sensitivity analysis showed smaller and more uncertain effects (ESM Tables 6, 7). Glycaemic variability calculated as CV_{CGM} likewise showed smaller effect sizes for the dapagliflozin and exercise groups and no effects in the metformin group compared with control (Table 2). Compared with metformin, MAGE was reduced by 17.2% in the dapagliflozin group, but with no differences between the effects in the exercise and dapagliflozin groups (Fig. 2, Table 2). Both the dapagliflozin and exercise groups experienced a reduction in MAGE from baseline to 6 weeks and from baseline to 26 weeks compared with the control group (Fig. 2, Table 2). For details on the distribution of CGM-based measures see ESM Figs 2–7.

Secondary outcomes

Other glycaemic measures In all three active groups, a mean decline in HbA_{1c} of ~1 mmol/mol (~0.1%) was observed at 13 weeks compared with control (Table 3). At 26 weeks, HbA_{1c} had returned to the baseline level in all groups. Mean fasting plasma glucose concentration decreased by 0.3 mmol/l in the metformin group at 13 weeks (Table 3). In the same period, the OGTT $iAUC_{glucose}$ increased by ~40% in the metformin group compared with the control group; this increase was still present at 26 weeks (Table 3). At 13 weeks, a 20–24% decline was observed in the fasting serum insulin

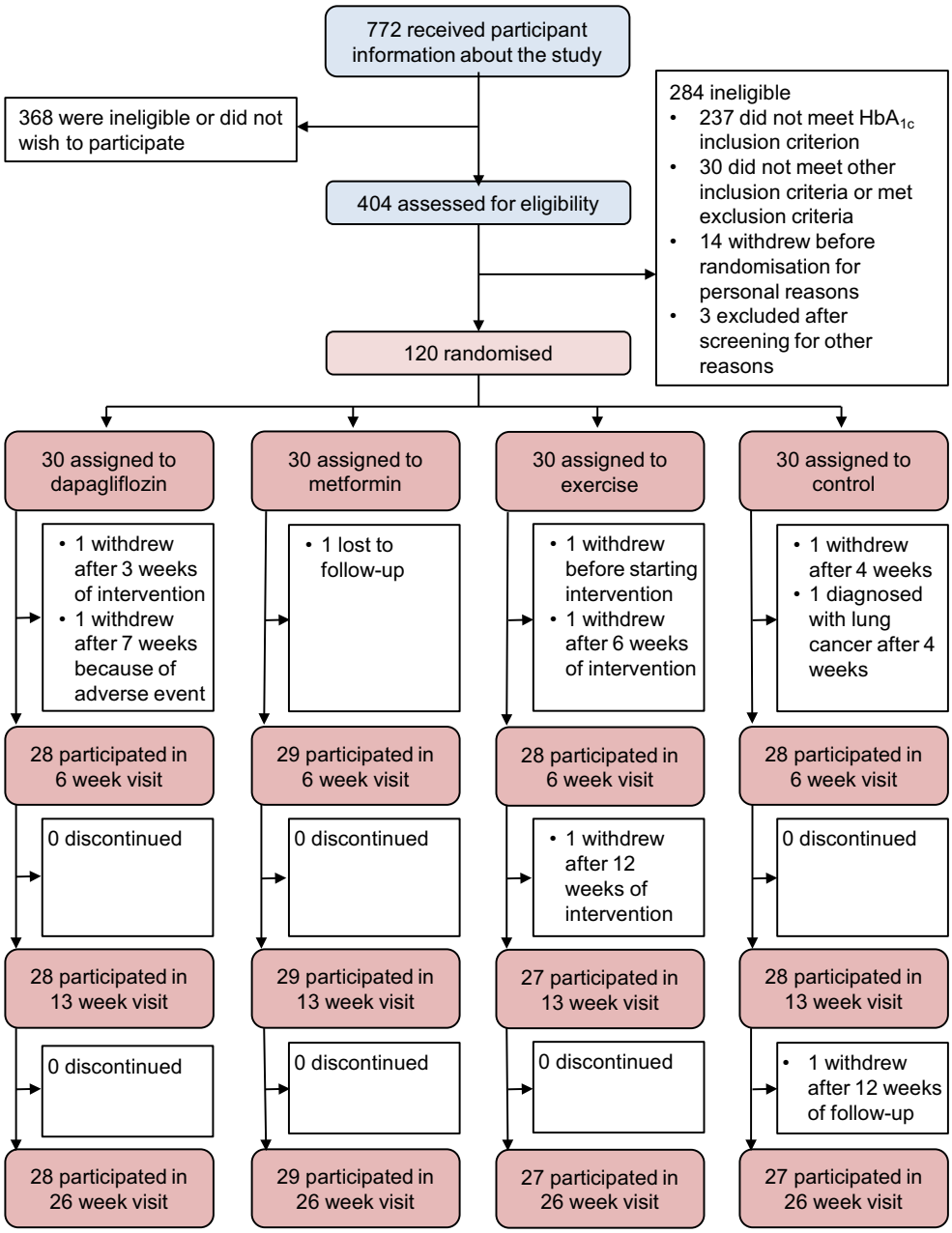

Fig. 1 Trial profile

concentration in the metformin and dapagliflozin groups; these changes were maintained at the 26 week follow-up. The OGTT $iAUC_{insulin}$ decreased by 20% in the dapagliflozin group and by 18% in the exercise group but was unchanged in the metformin group at 13 weeks. Participants in the exercise group reduced their time spent at >7.0 mmol/l when compared with those in the control group at 13 weeks (Table 3). At baseline, few participants spent a significant amount of time with a sensor glucose level of >7.8 or >11.1 mmol/l (Table 1) and these outcomes were consequently not analysed further.

Anthropometric measures Reductions in body weight of ~1 kg were observed in the dapagliflozin and metformin groups at 6 and 13 weeks; however, greater variation was observed at 13 weeks (Table 4). At 13 weeks, participants in the exercise group had on average reduced their body weight by 1.4 kg (~1.2%) compared with control. This change was accompanied by mean reductions of 2.5 cm in waist circumference and 1.1 kg in fat mass. Individuals randomised to dapagliflozin had on average reduced their waist circumference by 2.4 cm. Fat-free mass was unaltered in all three treatment groups compared with control (Table 4).

Markers of cardiometabolic function None of the three treatments were associated with changes in BP compared with control (Table 5). A mean reduction in LDL-cholesterol

Table 2 Pairwise comparisons of the change in the primary outcome (MAGE) and other measures of glycaemic variability from baseline to 6, 13 and 26 weeks

Measure	Week 6		Week 13		Week 26	
	Difference	*p* value	Difference	*p* value	Difference	*p* value
MAGE						
DAP vs CON	−16.6 (−30.3, −0.2)	0.047	−17.1 (−30.8, −0.7)	0.042	−18.0 (−31.7, −1.7)	0.032
MET vs CON	−7.6 (−22.4, 10.2)	0.379	0.1 (−16.1, 19.4)	0.991	−9.1 (−23.9, 8.5)	0.291
EXE vs CON	−15.7 (−29.3, 0.6)	0.059	−15.3 (−29.1, 1.2)	0.067	−21.4 (−34.5, −5.7)	0.009
DAP vs MET	−9.8 (−24.5, 7.8)	0.257	−17.2 (−30.9, −0.8)	0.041	−9.8 (−24.6, 7.8)	0.256
EXE vs DAP	1.1 (−15.5, 20.9)	0.903	2.2 (−14.8, 22.5)	0.815	−4.1 (−20.2, 15.1)	0.651
EXE vs MET	−8.8 (−23.4, 8.7)	0.305	−15.4 (−29.1, 1.1)	0.065	−13.6 (−27.7, 3.4)	0.110
SD						
DAP vs CON	−13.3 (−26.2, 1.9)	0.083	−15.8 (−28.4, −0.9)	0.039	−12.1 (−25.4, 3.6)	0.124
MET vs CON	−7.6 (−21.1, 8.2)	0.324	−0.3 (−15.0, 16.8)	0.966	−7.2 (−20.9, 8.9)	0.362
EXE vs CON	−9.6 (−22.9, 6.0)	0.215	−9.1 (−22.5, 6.7)	0.245	−12.5 (−25.7, 3.1)	0.111
DAP vs MET	−6.1 (−20.0, 10.2)	0.438	−15.5 (−28.2, −0.5)	0.043	−5.3 (−19.3, 11.3)	0.509
EXE vs DAP	4.4 (−11.2, 22.5)	0.607	8.0 (−8.3, 27.2)	0.360	−0.5 (−15.6, 17.4)	0.956
EXE vs MET	−2.1 (−16.4, 14.7)	0.794	−8.8 (−22.2, 7.1)	0.262	−5.7 (−19.7, 10.7)	0.474
CV						
DAP vs CON	−10.0 (−22.8, 4.8)	0.176	−13.4 (−25.7, 1.1)	0.068	−11.5 (−24.2, 3.3)	0.121
MET vs CON	−4.6 (−17.9, 10.7)	0.534	−1.2 (−15.0, 14.8)	0.870	−7.3 (−20.3, 7.9)	0.329
EXE vs CON	−8.2 (−21.0, 6.7)	0.267	−6.1 (−19.3, 9.2)	0.414	−11.1 (−23.8, 3.9)	0.139
DAP vs MET	−5.6 (−18.9, 9.8)	0.454	−12.3 (−24.8, 2.3)	0.096	−4.6 (−18.1, 11.1)	0.543
EXE vs DAP	2.0 (−12.4, 18.9)	0.795	8.3 (−7.2, 26.6)	0.310	0.5 (−14.0, 17.5)	0.946
EXE vs MET	−3.7 (−17.1, 11.9)	0.621	−4.9 (−18.3, 10.6)	0.512	−4.1 (−17.6, 11.7)	0.590

Data were $\log_e$-transformed for analysis and back-transformed for presentation. Results are presented as relative changes and were calculated as the ratio of mean (95% CI) change from baseline in one group vs the other in percentage term (i.e. if group A changed 20% between visit 1 and 2 [A2/A1 = 1.2] and group B changed 10% [B2/B1 = 1.1], then the relative change between group A and B is 1.2/1.1 = 9.1%)

CON, control; DAP, dapagliflozin; EXE, exercise; MET, metformin

(−0.3 mmol/l) was observed in the metformin and exercise groups after 6 weeks, but at 13 and 26 weeks these changes were no longer apparent (Table 5). Exercise was associated with reductions in VLDL-cholesterol and triacylglycerol concentrations by 17–19% after 13 weeks (Table 5). From baseline to 13 weeks, participants in the exercise group increased CRF ($\dot{V}O_{2peak}$) by 2.8 ml O_2 min^{-1}; kg^{-1}, corresponding to an 11% increase, and decreased their resting heart rate by 4 beats/min compared with those in the control group; these changes were still present at 26 weeks (Table 5).

Adverse events

One serious adverse event occurred in the control group (lung cancer). During the active intervention period (baseline to 13 weeks), 44% of the participants experienced at least one adverse event (dapagliflozin 57%, metformin 80%, exercise 23%, control 17%). Most adverse events were known side-effects of the two medications; in the exercise group most events were related to the musculoskeletal system (Fig. 3 and ESM Results, ESM Tables 8, 9).

Discussion

We found that 13 weeks of dapagliflozin treatment reduced glycaemic variability (MAGE) compared with control or treatment with metformin in individuals with overweight and prediabetes diagnosed by HbA_{1c}. A similar effect was observed for interval-based exercise, though with a slightly smaller effect size and greater uncertainty of the estimate. However, the effect sizes did not reach the predefined minimal clinically important difference, and were even smaller when reported as CV_{CGM}, which is now recommended as the primary measure of glycaemic variability [19]. The lack of clinically relevant effects was supported by the analysis based on per protocol completers and by the post hoc analysis with adjustment for the baseline level of MAGE. Dapagliflozin, metformin and exercise were associated with reductions in HbA_{1c} and improved markers of

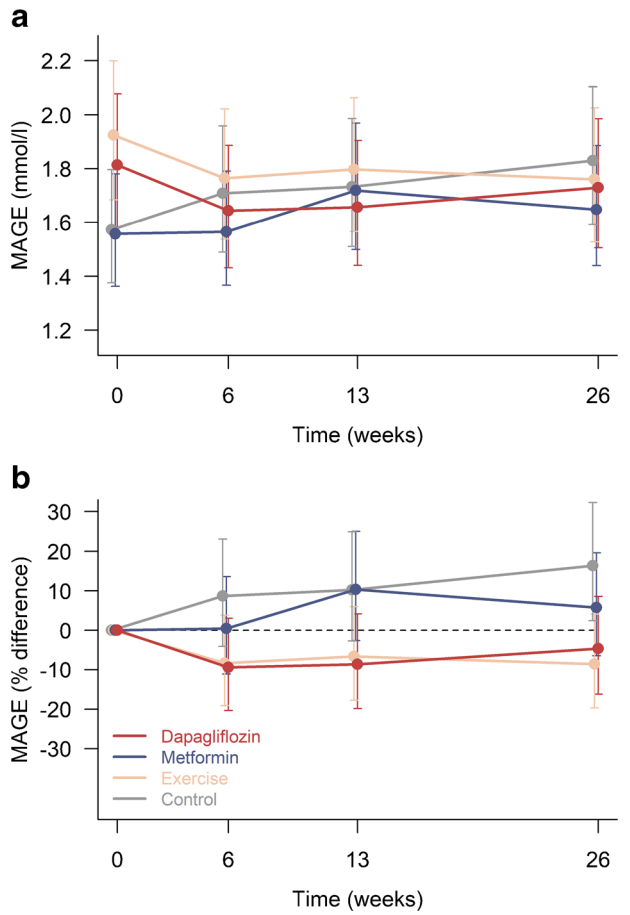

Fig. 2 Intervention effects for the primary outcome, MAGE. Results were back-transformed for presentation and are presented as estimated conditional geometric means (95% CI) (**a**) and estimated relative changes from baseline (95% CI) (**b**)

glucometabolic function; however, we observed an increase in post-load glucose levels in the metformin group. No major adverse events were recorded in the active treatment groups, and the exercise group experienced fewest events.

Raised fasting and OGTT-derived glucose concentrations as well as mean glucose levels are well-established risk factors for the development of diabetes, CVD and premature death [2, 29]. At the same time, glycaemic variability is emerging as an independent risk factor [18, 19, 30]. This is the first trial to assess the efficacy of SGLT2 inhibition on glycaemic variability in people with prediabetes diagnosed by HbA_{1c}. We compared SGLT2 inhibition not only with a control group but also head-to-head with treatments that have proven efficacious in preventing diabetes in people with impaired glucose tolerance [3–5]. As we hypothesised, treatment with dapagliflozin or interval-based exercise was superior to metformin in improving glycaemic variability when measured in the week after the end of treatment. However, the effect sizes observed for MAGE were around half the size of what we expected and the variation in the effect estimates was large. In contrast to what we expected, exercise was not superior to

Table 3 Change in secondary glycaemic outcomes between the three active groups and the control group from baseline to 6, 13 and 26 weeks

Outcome	Week 6	Week 13	Week 26
HbA_{1c}, mmol/mol			
DAP	−1.2 (−2.1, −0.4)	−1.3 (−2.3, −0.3)	0.0 (−0.9, 0.9)
MET	0.1 (−0.7, 0.9)	−1.2 (−2.3, −0.2)	0.4 (−0.5, 1.3)
EXE	−0.7 (−1.5, 0.1)	−1.1 (−2.1, 0.0)	0.2 (−0.7, 1.1)
HbA_{1c}, %			
DAP	−0.1 (−0.2, −0.0)	−0.1 (−0.2, −0.0)	0.0 (−0.1, 0.1)
MET	0.0 (−0.1, 0.1)	−0.1 (−0.2, −0.0)	0.0 (−0.1, 0.1)
EXE	−0.1 (−0.1, 0.0)	−0.1 (−0.2, 0.0)	0.0 (−0.1, 0.1)
Fasting plasma glucose, mmol/l			
DAP	−0.3 (−0.6, 0.0)	−0.1 (−0.4, 0.1)	0.0 (−0.3, 0.2)
MET	−0.5 (−0.8, −0.2)	−0.3 (−0.6, −0.1)	0.0 (−0.3, 0.3)
EXE	−0.2 (−0.5, 0.1)	−0.1 (−0.3, 0.1)	0.0 (−0.3, 0.3)
Fasting serum insulin, %[a]			
DAP	−19 (−37, 4)	−24 (−39, −6)	−26 (−44, −2)
MET	−23 (−40, −1)	−20 (−35, −1)	−22 (−41, 3)
EXE	−25 (−42, −4)	−15 (−32, 5)	−14 (−35, 13)
$iAUC_{glucose}$, mmol/l × min			
DAP		−29 (−93, 35)	7 (−61, 75)
MET		112 (48, 176)	80 (12, 148)
EXE		3 (−62, 68)	−42 (−111, 26)
$iAUC_{insulin}$, %[a]			
DAP		−20 (−35, −1)	−11 (−26, 8)
MET		−1 (−20, 22)	0 (−17, 20)
EXE		−18 (−34, 1)	−14 (−29, 4)
Mean $glucose_{CGM}$, mmol/l			
DAP	−0.2 (−0.5, 0.0)	−0.2 (−0.4, 0.0)	0.0 (−0.3, 0.2)
MET	−0.2 (−0.4, 0.0)	0.0 (−0.2, 0.3)	0.0 (−0.2, 0.2)
EXE	−0.1 (−0.3, 0.1)	−0.2 (−0.4, 0.0)	−0.1 (−0.3, 0.1)
Percentage of time spent at >6.1 mmol/l[a]			
DAP	−21 (−60, 56)	−13 (−56, 74)	−13 (−57, 73)
MET	18 (−40, 130)	80 (−8, 253)	55 (−21, 205)
EXE	−3 (−51, 90)	−46 (−73, 5)	−10 (−55, 81)
Percentage of time spent at >7.0 mmol/l %[a]			
DAP	−64 (−87, 1)	−55 (−84, 27)	−44 (−80, 59)
MET	40 (−49, 283)	98 (−28, 445)	51 (−46, 318)
EXE	−28 (−74, 100)	−66 (−88, −5)	−49 (−82, 47)

Data are estimated mean changes (95% CI) from baseline compared with CON

Results for the transformed outcomes are presented as relative changes and were calculated as the ratio of mean (95% CI) change from baseline in one group vs the other in percentage term (i.e. if group A changed 20% between visit 1 and 2 [A2/A1 = 1.2] and group B changed 10% [B2/B1 = 1.1], then the relative change between group A and B is 1.2/1.1 = 9.1%)

[a] Ln-transformed before analysis and back-transformed for presentation

CON, control; DAP, dapagliflozin; EXE, exercise; MET, metformin

treatment with dapagliflozin in reducing glycaemic variability, potentially because the exercise intensity in our study was too

Table 4 Change in anthropometric measures between the three active groups and the control group from baseline to 6, 13 and 26 weeks

Outcome	Week 6	Week 13	Week 26
Weight, kg[a]			
DAP	−1.3 (−2.3, −0.4)	−1.1 (−2.6, 0.3)	−0.9 (−2.9, 1.2)
MET	−0.9 (−1.8, 0.1)	−0.9 (−2.3, 0.6)	−1.3 (−3.3, 0.8)
EXE	−0.6 (−1.5, 0.4)	−1.4 (−2.8, 0.1)	−1.5 (−3.5, 0.6)
BMI, kg/m^2			
DAP	−0.4 (−0.7, −0.1)	−0.3 (−0.8, 0.1)	−0.3 (−0.9, 0.4)
MET	−0.3 (−0.6, 0.1)	−0.3 (−0.7, 0.2)	−0.4 (−1.0, 0.3)
EXE	−0.2 (−0.5, 0.1)	−0.4 (−0.9, 0.0)	−0.4 (−1.1, 0.3)
WC, cm[a]			
DAP	−1.9 (−4.1, 0.2)	−2.4 (−4.8, 0.0)	−2.3 (−5.3, 0.7)
MET	−1.5 (−3.7, 0.7)	−0.8 (−3.2, 1.6)	−2.1 (−5.1, 0.9)
EXE	−1.8 (−4.0, 0.4)	−2.5 (−5.0, −0.1)	−1.2 (−4.3, 1.9)
WHR[a]			
DAP	0.00 (−0.03, 0.02)	−0.02 (−0.04, 0.01)	−0.02 (−0.05, 0.00)
MET	0.00 (−0.03, 0.02)	−0.01 (−0.04, 0.01)	−0.02 (−0.05, 0.00)
EXE	0.00 (−0.02, 0.03)	−0.03 (−0.05, 0.00)	−0.02 (−0.05, 0.01)
Fat mass, kg[a]			
DAP		−0.7 (−1.8, 0.3)	−0.4 (−2.0, 1.2)
MET		−0.7 (−1.7, 0.3)	−0.7 (−2.3, 0.8)
EXE		−1.1 (−2.1, 0.0)	−0.9 (−2.4, 0.7)
Fat-free mass, kg[a]			
DAP		−0.4 (−1.3, 0.4)	−0.7 (−1.7, 0.2)
MET		−0.2 (−1.0, 0.7)	−0.4 (−1.4, 0.5)
EXE		−0.1 (−1.0, 0.8)	−0.4 (−1.4, 0.6)

Data are estimated mean changes (95% CI) from baseline compared with control

[a] Adjusted for sex

CON, control; DAP, dapagliflozin; EXE, exercise; MET, metformin; WC, waist circumference

low to result in improvements in glycaemic variability of the magnitude observed in other studies [20, 21].

The PRE-D Trial was designed to investigate on-treatment effects on glycaemic variability (6 weeks) in addition to assessing both short-term (13 weeks) and longer-term (26 weeks) physiological post-treatment adaptations. Interestingly, small improvements in MAGE in the dapagliflozin group were observed during treatment, immediately after treatment and at the 26 week follow-up. Whether this longer-term effect represents an actual physiological legacy effect or whether it is a random finding is too early to conclude. An effect on glycaemic variability in the exercise group was also observed at the follow-up visit at 26 weeks. Support for the exercise programme was withdrawn at 13 weeks but the improvements in CRF and resting heart rate at 26 weeks indicate that the participants continued their

Table 5 Change in markers of cardiometabolic function between the three active groups and the control group from baseline to 6, 13 and 26 weeks

Outcome	Week 6	Week 13	Week 26
Systolic BP, mmHg			
DAP	−4 (−10, 2)	−3 (−9, 4)	1 (−6, 8)
MET	−1 (−8, 5)	−1 (−8, 5)	2 (−5, 10)
EXE	3 (−4, 9)	1 (−5, 8)	3 (−4, 10)
Diastolic BP, mmHg			
DAP	−2 (−6, 1)	0 (−5, 4)	−1 (−5, 3)
MET	0 (−4, 4)	1 (−4, 5)	−2 (−6, 1)
EXE	−2 (−5, 2)	3 (−2, 7)	−1 (−5, 2)
Resting heart rate, bpm			
DAP	−2 (−6, 2)	0 (−5, 4)	−3 (−8, 2)
MET	1 (−3, 6)	2 (−2, 6)	−2 (−8, 3)
EXE	−5 (−10, −1)	−4 (−8, 0)	−5 (−11, 0)
CRF, ml O_2 min^{-1}[a] kg^{-1}			
DAP		0.9 (−0.6, 2.3)	0.4 (−1.4, 2.3)
MET		−0.3 (−1.7, 1.2)	1.4 (−0.4, 3.3)
EXE		2.8 (1.3, 4.2)	2.9 (1.1, 4.8)
Total cholesterol, mmol/l			
DAP	0.0 (−0.4, 0.3)	0.0 (−0.4, 0.3)	−0.3 (−0.7, 0.2)
MET	−0.3 (−0.6, 0.0)	−0.2 (−0.5, 0.2)	−0.2 (−0.7, 0.2)
EXE	−0.3 (−0.6, 0.0)	−0.1 (−0.4, 0.3)	−0.2 (−0.6, 0.3)
LDL-cholesterol, mmol/l			
DAP	−0.1 (−0.3, 0.2)	−0.1 (−0.4, 0.2)	−0.3 (−0.6, 0.1)
MET	−0.3 (−0.5, 0.0)	−0.2 (−0.5, 0.1)	−0.2 (−0.5, 0.2)
EXE	−0.3 (−0.5, 0.0)	−0.1 (−0.4, 0.2)	−0.1 (−0.5, 0.2)
HDL-cholesterol, mmol/l[a]			
DAP	0.08 (−0.02, 0.19)	0.15 (0.04, 0.25)	0.04 (−0.08, 0.16)
MET	−0.06 (−0.17, 0.04)	0.00 (−0.11, 0.11)	−0.04 (−0.17, 0.08)
EXE	0.04 (−0.07, 0.14)	0.08 (−0.02, 0.19)	0.01 (−0.12, 0.13)
VLDL-cholesterol, %[b]			
DAP	−5 (−19, 12)	−13 (−27, 4)	−5 (−20, 14)
MET	10 (−6, 30)	−5 (−20, 13)	−11 (−26, 6)
EXE	−6 (−21, 11)	−17 (−30, −1)	−11 (−26, 7)
Triacylglycerols, %[b]			
DAP	−6 (−21, 10)	−12 (−26, 5)	−4 (−20, 15)
MET	7 (−9, 26)	−1 (−17, 17)	−9 (−25, 9)
EXE	−9 (−23, 7)	−19 (−32, −3)	−15 (−29, 3)

Data are estimated mean changes (95% CI) from baseline compared with control. The results for the transformed outcomes are presented as relative changes and were calculated as the ratio of mean (95% CI) change from baseline in one group vs the other in percentage term (i.e. if group A changed 20% between visit 1 and 2 [A2/A1 = 1.2] and group B changed 10% [B2/B1 = 1.1], then the relative change between group A and B is 1.2/1.1 = 9.1%)

[a] Adjusted for sex

[b] Ln-transformed before analysis and back-transformed for presentation

CON, control; DAP, dapagliflozin; EXE, exercise; MET, metformin

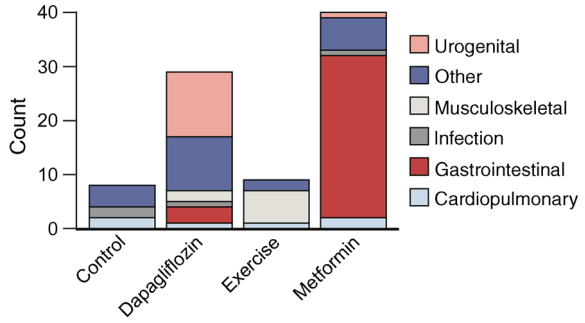

Fig. 3 The number of adverse events reported within each group from baseline to end of treatment (13 weeks) presented by organ system

exercise in the period between the last two visits, which could explain the beneficial effects on the glucometabolic outcomes observed at the end of the follow-up period in the exercise group.

We consider the use of a primary outcome obtained under 'free-living' conditions to be a major strength. Still, while this enables the evaluation of intervention effects in everyday life, it also poses a challenge as the clinical implications of CGM-derived measures, such as MAGE and CV_{CGM}, are still unclear [18] and relevant effect sizes are unknown, particularly in a population with prediabetes. Furthermore, the use of CGM as a primary outcome is challenging because of lack of consensus in the data management process, making comparisons across trials difficult. During the period in which the trial was running, CV_{CGM} was suggested as the preferred measure of glycaemic variability in diabetes [19]; this will hopefully improve and optimise comparison between studies using CGM data in the future. Our results indicate, however, that if CV_{CGM} had been the primary outcome of the trial, the effect sizes would have been even smaller. The small effect sizes for MAGE are potentially explained by the poor overlap between HbA_{1c} and glucose levels in the prediabetic range [9, 10], resulting in relatively low baseline levels of MAGE compared with observations made in individuals with impaired fasting glucose or impaired glucose tolerance [30, 31]. Accordingly, the potential for improvement in MAGE in our study population was modest. Furthermore, the baseline MAGE levels in the control and metformin groups were relatively low, which could theoretically lead to a floor effect in these groups.

We included overweight or obese participants with prediabetes defined solely by HbA_{1c}. This population is easily identified in the clinical setting compared with prediabetes defined by impaired fasting glucose or impaired glucose tolerance, but evidence for the optimal glucose-lowering strategy in this high-risk group has been lacking. Despite between-group variations in glycaemic variability, high inter-individual variation in post-load glucose and insulin levels, and relatively normal glycaemic control at baseline, all active treatments were associated with improvements in some of the secondary glucometabolic outcomes. Clinically relevant effect sizes for the glycaemic outcomes are unknown for this population and although we only observed small improvements in blood glucose levels, the size of the improvements in HbA_{1c} were comparable to those obtained in the Diabetes Prevention Program [3] and could thus hypothetically have substantial long-term effects, especially when considering the marked improvements in both fasting and post-load insulin levels. This underlines the need for long-term diabetes prevention trials in individuals with prediabetes defined by HbA_{1c}. However, the normal fasting and/or 2 h glucose levels exhibited by many of our participants at baseline indicate that individuals with prediabetic HbA_{1c} levels in the low end of the range suggested by the ADA (39–47 mmol/mol [5.7–6.4%]) [17] may not be the optimal target group for interventions aimed at preventing diabetes. Potentially, the cut-points suggested by the International Expert Committee (42–47 mmol/mol [6.0–6.4%]) [32] may be more relevant for diabetes prevention efforts. Nevertheless, because HbA_{1c} is closely associated with cardiovascular risk in both the non-diabetic and diabetic range [33], assessing the long-term effects of interventions targeting cardiometabolic risk (e.g. diet, exercise and/or SGLT2 inhibition [34, 35]) are still relevant for all individuals with prediabetes defined by the ADA's criteria.

Moving beyond glucose metabolism, the interventions were associated with improvement in other markers of cardiometabolic function. Treatment with either dapagliflozin or exercise was associated with small improvements in body weight and waist circumference. In the dapagliflozin group, part of the initial weight loss was likely secondary to some degree of dehydration [13], as the weight loss was not accompanied by a loss of fat mass. Furthermore, the weight loss had diminished at 13 weeks. In the exercise group, the weight loss seemed primarily to be driven by a loss of fat mass. Exercise was also associated with improvement in CRF and decreases in plasma triacylglycerol levels; these are well-known beneficial effects of physical activity [36], representing improvements beyond those attained by the two pharmacological treatments. An increase in CRF similar to that observed in the exercise group has been proposed to be relevant for reducing the risk of CVD in a sedentary population [37].

In conclusion, treatment with dapagliflozin or interval-based exercise improved glycaemic variability compared head-to-head with habitual lifestyle or metformin therapy in individuals with prediabetes diagnosed by HbA_{1c} but the effect sizes were small and likely of limited clinical relevance. All treatments were associated with small improvements in glucometabolic outcomes, and exercise was associated with additional cardiometabolic benefits. Given the risk of adverse events and limited effects on blood glucose levels, our results do not lend support to the benefits of the pharmacological interventions in the short-term outweighing the harms in this population with prediabetes, especially not metformin treatment. Additionally, our findings

raise the question of whether individuals with HbA_{1c} levels in the lower prediabetic range, as specified by the ADA, are ideal targets for pharmaceutical glucose-lowering interventions with metformin or SGLT2 inhibitors.

Acknowledgements We are grateful to the study participants for their cooperation and willingness to participate. The laboratory technicians H. Vishof, L. S. Koch, S. Sidenius and C. S. Nielsen at Steno Diabetes Center Copenhagen (SDCC) are thanked for their great dedication, skilled assistance and coordination. C. S. Hansen, N. Safai, N. J. Johansen, J. I. B. Egholk (SDCC) are thanked for their assistance during the study. This work was supported by the Novo Nordisk Foundation, AstraZeneca AB, the Danish Innovation Foundation and University of Copenhagen. Parts of this study were presented in abstract form at the 79th Scientific Sessions of the ADA, San Francisco, CA, 7–11 June 2019 and at the 55th Annual Meeting of the EASD, Barcelona, 16–20 September 2019.

Data availability The datasets generated during and/or analysed during the current study are available from the corresponding author on reasonable request.

Funding The study was funded by the Novo Nordisk Foundation, AstraZeneca AB, the Danish Innovation Foundation, the University of Copenhagen and Ascensia Diabetes Care Denmark ApS. The funders had no role in study design, data collection, data analysis, interpretation or writing of the report.

Authors' relationships and activities All authors have completed the ICMJE uniform disclosure form (available on request from the corresponding author) and declare that KF, HA, LB, DV, FP, KKBC, MBB, CTRV, MT, TFD and MEJ are employed by Steno Diabetes Center Copenhagen, a research hospital working in the Danish National Health Service. Until 31 December 2016 Steno Diabetes Center was owned by Novo Nordisk A/S and received part of its core funding from unrestricted grants from the Novo Nordisk Foundation and Novo Nordisk A/S. KF, DV and MEJ own shares in Novo Nordisk A/S. KF has received research support from AstraZeneca. TFD has consulted for Novo Nordisk, AstraZeneca and Boehringer Ingelheim and has received research support from Novo Nordisk and AstraZeneca. MR-L has received personal lecture fees from Novo Nordisk A/S. FP reports having received research grants from AstraZeneca, Novo Nordisk and Novartis and lecture fees from MSD, AstraZeneca, Novo Nordisk, Novartis, Eli Lilly and Boehringer Ingelheim and has served as a consultant for AstraZeneca, Novo Nordisk, Amgen and MSD. MEJ has received research grants from Amgen, Sanofi Aventis, Boehringer Ingelheim and Astra Zeneca.

Contribution statement KF, FP and MEJ conceived the idea and designed the study. KF is the sponsor and MEJ is the principal investigator. LB, MR-L and KK contributed to the design of the exercise intervention. KF, LB, HA, MBB, CTRV, CP, KKBC, MT, TFD and MEJ were involved in the conduct of the trial and data collection. KKBC, MBB and DV performed statistical analyses. KF and MBB drafted the manuscript. All authors critically revised the manuscript for important intellectual content and approved the final version of the manuscript. KF is the guarantor of this work and, as such, had full access to all the data in the study, takes responsibility for the integrity of the data and the accuracy of the data analysis, and had final responsibility for the decision to submit for publication.

References

1. Eckel RH, Kahn SE, Ferrannini E et al (2011) Obesity and type 2 diabetes: what can be unified and what needs to be individualized? J Clin Endocrinol Metab 96(6):1654–1663. https://doi.org/10.1210/jc.2011-0585
2. Færch K, Johansen NB, Vistisen D, Jørgensen ME (2014) Cardiovascular risk stratification and management in pre-diabetes. Curr Diab Rep 14:493
3. Knowler WC, Barrett-Connor E, Fowler SE et al (2002) Reduction in the incidence of type 2 diabetes with lifestyle intervention or metformin. N Engl J Med 346(6):393–403
4. Tuomilehto J, Lindström J, Eriksson JG et al (2001) Prevention of type 2 diabetes mellitus by changes in lifestyle among subjects with impaired glucose tolerance. N Engl J Med 344(18):1343–1350
5. Pan XR, Li GW, Hu YH et al (1997) Effects of diet and exercise in preventing NIDDM in people with impaired glucose tolerance. The Da Qing IGT and Diabetes Study. Diabetes Care 20(4):537–544
6. Saito T, Watanabe M, Nishida J et al (2011) Lifestyle modification and prevention of type 2 diabetes in overweight Japanese with impaired fasting glucose levels: a randomized controlled trial. Arch Intern Med 171(15):1352–1360
7. Færch K, Hulman A, Solomon TP (2015) Heterogeneity of pre-diabetes and type 2 diabetes: implications for prediction, prevention and treatment responsiveness. Curr Diab Rev 12(1):30–41
8. American Diabetes Association (2019) Prevention or delay of type 2 diabetes: standards of medical care in diabetes-2019. Diabetes Care 42(Suppl 1):S29–S33. https://doi.org/10.2337/dc19-S003
9. Færch K, Johansen NB, Witte DR, Lauritzen T, Jørgensen ME, Vistisen D (2015) Relationship between insulin resistance and β-cell dysfunction in subphenotypes of prediabetes and type 2 diabetes. J Clin Endocrinol Metab 100(2):707–716. https://doi.org/10.1210/jc.2014-2853
10. Barry E, Roberts S, Oke J, Vijayaraghavan S, Normansell R, Greenhalgh T (2017) Efficacy and effectiveness of screen and treat policies in prevention of type 2 diabetes: systematic review and meta-analysis of screening tests and interventions. BMJ 356: i6538. https://doi.org/10.1136/bmj.i6538
11. Færch K, Borch-Johnsen K, Vaag A, Jørgensen T, Witte D (2010) Sex differences in glucose levels: a consequence of physiology or methodological convenience? The Inter99 study. Diabetologia 53(5):858–865
12. Xu Y, Wang L, He J et al (2013) Prevalence and control of diabetes in Chinese adults. JAMA 310(9):948–959. https://doi.org/10.1001/jama.2013.168118
13. Lee PC, Ganguly S, Goh SY (2018) Weight loss associated with sodium-glucose cotransporter-2 inhibition: a review of evidence and underlying mechanisms. Obes Rev 19(12):1630–1641. https://doi.org/10.1111/obr.12755
14. Ramírez Rodríguez AM, González Ortiz M, Martínez Abundis E (2020) Effect of dapagliflozin on insulin secretion and insulin sensitivity in patients with prediabetes. Exp Clin Endocrinol Diabetes 128(8):506–511
15. Al Jobori H, Daniele G, Adams J et al (2017) Determinants of the increase in ketone concentration during SGLT2 inhibition in NGT, IFG and T2DM patients. Diabetes Obes Metab 19(6):809–813. https://doi.org/10.1111/dom.12881
16. Ferrannini E, Baldi S, Frascerra S et al (2016) Shift to fatty substrate utilization in response to sodium-glucose cotransporter 2 inhibition in subjects without diabetes and patients with type 2 diabetes. Diabetes 65(5):1190–1195. https://doi.org/10.2337/db15-1356
17. American Diabetes Association (2019) Classification and diagnosis of diabetes: standards of medical care in diabetes-2019. Diabetes Care 42(Suppl 1):S13–S28. https://doi.org/10.2337/dc19-S002

18. Ceriello A, Monnier L, Owens D (2019) Glycaemic variability in diabetes: clinical and therapeutic implications. Lancet Diabetes Endocrinol 7(3):221–230. https://doi.org/10.1016/s2213-8587(18)30136-0
19. Danne T, Nimri R, Battelino T et al (2017) International consensus on use of continuous glucose monitoring. Diabetes Care 40(12):1631–1640. https://doi.org/10.2337/dc17-1600
20. Karstoft K, Clark MA, Jakobsen I et al (2017) The effects of 2 weeks of interval vs continuous walking training on glycaemic control and whole-body oxidative stress in individuals with type 2 diabetes: a controlled, randomised, crossover trial. Diabetologia 60(3):508–517. https://doi.org/10.1007/s00125-016-4170-6
21. Rafiei H, Robinson E, Barry J, Jung M, Little J (2019) Short-term exercise training reduces glycaemic variability and lowers circulating endothelial microparticles in overweight and obese women at elevated risk of type 2 diabetes. Eur J Sport Sci 19(8):1140–1149. https://doi.org/10.1080/17461391.2019.1576772
22. Færch K, Amadid H, Nielsen LB et al (2017) Protocol for a randomised controlled trial of the effect of dapagliflozin, metformin and exercise on glycaemic variability, body composition and cardiovascular risk in prediabetes (the PRE-D Trial). BMJ Open 7(5):e013802. https://doi.org/10.1136/bmjopen-2016-013802
23. American Diabetes Association (2017) Classification and diagnosis of diabetes. Diabetes Care 40(Suppl 1):S11–S24. https://doi.org/10.2337/dc17-S005
24. Ministry of Environment and Food of Denmark (2015) The Danish official dietary recommendations. Available from http://altomkost.dk/raad-og-anbefalinger/de-officielle-kostraad/. Accessed 3 Oct 2018
25. Service FJ, Molnar GD, Rosevear JW, Ackerman E, Gatewood LC, Taylor WF (1970) Mean amplitude of glycemic excursions, a measure of diabetic instability. Diabetes 19(9):644–655. https://doi.org/10.2337/diab.19.9.644
26. Friedewald WT, Levy RI, Fredrickson DS (1972) Estimation of the concentration of low-density lipoprotein cholesterol in plasma, without use of the preparative ultracentrifuge. Clin Chem 18(6):499–502
27. Levey AS, Stevens LA (2010) Estimating GFR using the CKD Epidemiology Collaboration (CKD-EPI) creatinine equation: more accurate GFR estimates, lower CKD prevalence estimates, and better risk predictions. Am J Kidney Dis 55(4):622–627. https://doi.org/10.1053/j.ajkd.2010.02.337
28. Battelino T, Danne T, Bergenstal RM et al (2019) Clinical targets for continuous glucose monitoring data interpretation: recommendations from the International Consensus on Time in Range. Diabetes Care 42(8):1593–1603. https://doi.org/10.2337/dci19-0028
29. Borg R, Kuenen JC, Carstensen B et al (2011) HbA_{1c} and mean blood glucose show stronger associations with cardiovascular disease risk factors than do postprandial glycaemia or glucose variability in persons with diabetes: the A1C-Derived Average Glucose (ADAG) study. Diabetologia 54(1):69–72. https://doi.org/10.1007/s00125-010-1918-2
30. Hanefeld M, Sulk S, Helbig M, Thomas A, Köhler C (2014) Differences in glycemic variability between normoglycemic and prediabetic subjects. J Diabetes Sci Technol 8(2):286–290
31. Chakarova N, Dimova R, Grozeva G, Tankova T (2019) Assessment of glucose variability in subjects with prediabetes. Diabetes Res Clin Pract 151:56–64. https://doi.org/10.1016/j.diabres.2019.03.038
32. The International Expert Committee (2009) International Expert Committee report on the role of the A1C assay in the diagnosis of diabetes. Diabetes Care 32(7):1327–1334
33. Selvin E, Steffes MW, Zhu H et al (2010) Glycated hemoglobin, diabetes, and cardiovascular risk in nondiabetic adults. N Engl J Med 362(9):800–811. https://doi.org/10.1056/NEJMoa0908359
34. Zinman B, Wanner C, Lachin JM et al (2015) Empagliflozin, cardiovascular outcomes, and mortality in type 2 diabetes. N Engl J Med 373(22):2117–2128. https://doi.org/10.1056/NEJMoa1504720
35. Perkovic V, Jardine MJ, Neal B et al (2019) Canagliflozin and renal outcomes in type 2 diabetes and nephropathy. N Engl J Med 380(24):2295–2306. https://doi.org/10.1056/NEJMoa1811744
36. Lin X, Zhang X, Guo J et al (2015) Effects of exercise training on cardiorespiratory fitness and biomarkers of cardiometabolic health: a systematic review and meta-analysis of randomized controlled trials. J Am Heart Assoc 4(7). https://doi.org/10.1161/JAHA.115.002014
37. Ross R, Blair SN, Arena R et al (2016) Importance of assessing cardiorespiratory fitness in clinical practice: a case for fitness as a clinical vital sign: a scientific statement from the American Heart Association. Circulation 134(24):e653–e699. https://doi.org/10.1161/CIR.0000000000000461