A systematic review examining the effects of sodium-glucose cotransporter-2 inhibitors (SGLT2is) on biomarkers of inflammation and oxidative stress

(Short title: SGLT2is, inflammation and oxidative stress in type 2 diabetes)

Jonathan J. H. Bray[1], Harri Foster-Davies[1], Jeffrey W. Stephens[1,2]

1. Department of Diabetes & Endocrinology, Morriston Hospital, Swansea, SA6 6NL
2. Diabetes Research Group, School of Medicine, Swansea University, Swansea SA2 8PP

Correspondence to:
Dr Jonathan J. H. Bray
Address: Department of Diabetes & Endocrinology, Morriston Hospital, Swansea, SA6 6NL
Email: jonathan.bray@doctors.org.uk

Word counts

Abstract: 196

Main manuscript: 4156

Number of tables: 2

Supplementary tables: 4

Number of figures: 2

Abstract

Aims

Sodium-glucose cotransporter-2 inhibitors (SGLT2is) have a protective cardiorenal effect in type 2 diabetes. This systematic review examines the effects of SGLT2is on clinical biomarkers of inflammation and oxidative stress.

Methods

A search of Medline, Embase, Web of Science, and The Cochrane Library was performed examining changes in selected clinical biomarkers for inflammation: c-reactive protein (CRP), adiponectin, interleukin-6 (IL6), tumour necrosis factor-alpha (TNF-α), and oxidative stress: 8-iso-prostaglandin F2α (8-iso-PGF2α) and 8-hydroxy-2'-deoxyguanosine (8-OHdG). Quality of evidence was evaluated using the GRADEpro tool and risk of bias was assessed using the Cochrane RoB 2 and ROBINS-I tools.

Results

A total of 23 (15 randomised, 8 observational) heterogeneously-designed clinical studies were identified (1,654 patients, 24 weeks median follow-up). Consistent reductions were observed for CRP (10/12 studies), IL6 (5/5 studies), TNFα (3/4 studies), 8-iso-PGF2α (3/4 studies) and 8-OHdG (2/2 studies), and a consistent increase in adiponectin (6/8 studies). Change in serum CRP following SGLT2is appear to be independent of change in HbA1c and other study design and clinically relevant variables.

Conclusions

There is heterogeneous, yet consistent data supporting the beneficial effects of SLGT2is on inflammatory and oxidative stress. Change in serum CRP appears to be independent of change in HbA1c.

Key words: Inflammation; oxidative stress, type 2 diabetes; sodium-glucose cotransporter-2 inhibitors.

Introduction

Type 2 diabetes mellitus (T2DM) is associated with considerable morbidity and mortality, with cardiovascular disease (CVD) accounting for over 50% of deaths (1) and diabetes-related kidney disease (DKD) accounting for up to 50% of all cases of end-stage renal failure (2). Furthermore, the risk of cardiovascular (CV) death is increased in the setting of T2DM with end-stage renal failure (3). CV mortality is due to a combination of atherosclerotic disease and heart failure (HF). Compared to patients without diabetes, those with T2DM are twice as likely to develop coronary artery disease (4) and 2-6 times more likely to have HF (5).

Sodium-glucose cotransporter-2 inhibitors (SGLT2is) have an established, yet unexplained cardiorenal protective effect (6-9). These agents act on sodium-glucose cotransporter-2 channels in the proximal convoluted tubule to promote glycosuria, and thereby improve glycaemic control and reduce glycated haemoglobin (HbA1c) (10). The major clinical trials investigating the effects of SGLT2is in relation to CV and renal outcomes include the Empagliflozin Cardiovascular Outcome Event Trial in Type 2 Diabetes Mellitus Patients (EMPA-REG); Canagliflozin Cardiovascular Assessment Study (CANVAS); the Multicenter Trial to Evaluate the Effect of Dapagliflozin on the Incidence of Cardiovascular Events (DECLARE-TIMI 58) and the Canagliflozin and Renal Outcomes in Type 2 Diabetes and Nephropathy (CREDENCE) trial (6-8, 11). A recent meta-analysis by Toyama et al, confirmed the benefits of SGLT2is (12). SGLT2is reduce the risk of hospitalisation or death due to HF by approximately 39%; decline in renal function by 29%; and 3-point major adverse CV events (MACE) (defined as CV death, non-fatal myocardial infarction or stroke) by 19%. The greatest benefits appear to be in reducing HF with a collective

reduction in relative risk of 39%. The CV benefits from SGLT2is are most apparent in patients with more advanced diabetes and established CV disease. This has been summarised in a meta-analysis by Zelniker et al (13). Indeed, patients without established CVD (but with CV risk factors) do not see a benefit in the 3-point MACE as opposed to patients with established CVD, where a 14% relative risk reduction is observed. SGLT2is also reduce the risk of renal composite outcomes by 33%, 44% and 56% in patients with an estimated glomerular filtration rate (eGFR) ≥90, 60-90 and <60 mL/min/m^3, respectively (13). This has now been taken into account in current guidelines, which recommended the use of SGLT2is in T2DM with established CVD, HF or chronic kidney disease (14).

SGLT2is appear to have pleiotropic effects in patients with T2DM that are unexplained by improved glycaemic control alone (15). T2DM is a condition associated with increased inflammation and oxidative stress (16, 17). Furthermore, inflammation is recognised in the pathogenesis of atherosclerosis (18) and DKD (19). Targeting inflammatory pathways have been shown to reduce the rate of recurrent cardiovascular events (20). Emerging evidence from basic scientific studies supports the view that SGLT2is may influence inflammation and oxidative stress (21-24) and that this may contribute to the improved outcomes associated with these agents. This systematic review aims to provide a comprehensive summary of the available clinical evidence examining the effects of SGLT2is on biomarkers of inflammation and oxidative stress.

Methods

Search Strategy

Medline, Embase, Web of Science, and the Cochrane Library were searched up to December 2019 using the following search terms:- (*gliflozin** OR *SGLT$2 inhibitor** OR *SGLT2 inhibitor** OR *sodium glucose cotransporter 2 inhibitor**) AND (*inflammation*/*oxidative stress* OR *biomarkers*). See PROSPERO (CRD42020180276) and supplementary table 5a and 5b for full search strategy. Where possible Mesh terms were used. This search strategy is illustrated in figure 1a. Two reviewers screened titles, and abstracts if necessary, to select clinical studies examining the effects of SGLT2is on inflammatory or oxidative stress biomarkers.

Published articles without original data including reviews, expert opinion, commentary and responses were excluded, along with research examining non-human or *in vitro* experiments. We included randomised controlled trials with a parallel and crossover design, along with observational studies. Both prospective and retrospective studies were included. All comparison designs were acceptable including studies comparing SGLT2i treated participants to placebo or standard diabetes therapies. Studies relating to canagliflozin, empagliflozin, dapagliflozin and luseogliflozin were included. There were no constraints applied to the use of concomitant antidiabetes medication, publication status, nor language. The study was conducted in-line with the Preferred Reporting Items for Systematic Reviews and Meta-Analyses (PRISMA) checklist. The selection process for publications is illustrated in figure 1b.

Selected biomarkers

A priori, we selected biomarkers widely accepted and reported in the scientific literature to be direct and effective indicators of inflammation and oxidative stress, these included inflammatory biomarkers: c-reactive protein (CRP), adiponectin, interleukin-6 (IL6), tumour necrosis factor-alpha (TNF-α), and oxidative stress biomarkers: 8-iso-prostaglandin F2α (8-iso-PGF2α) and 8-hydroxy-2'-deoxyguanosine (8-OHdG). Those with unproven validity or unclear significance in inflammation and oxidative stress were excluded.

Data extraction and quality assessment

Data was extracted to pre-formatted tables including study design, participant characteristics (table 1), and outcome measures. These data were used to produce a description of the serum inflammatory biomarker changes (table 2a) and changes in biomarkers of oxidative stress (table 2b). Original raw data extracted from studies can be found in supplementary table 1a and 1b. The strength of evidence was appraised using the Grading of Recommendations Assessment, Development and Evaluation (GRADEpro) tool (25) (supplementary table 2). Study quality was assessed using the Cochrane Risk-of-Bias Tool for Randomised trials (RoB 2) tool (26) and the Risk of bias in non-randomised studies of interventions (ROBINS-I) tool (27) (supplementary table 3a and 3b).

Associations with change in c-reactive protein (CRP)

We chose *a priori* to examine the relationship between change in inflammatory markers and mean blood glucose (HbA1c), study design factors (participant co-morbidity, pharmacological intensity, study follow-up period and SGLT2i concentration) and clinically relevant variables (eGFR, body mass index (BMI) and

homeostatic model assessment of insulin resistance (HOMA-IR)). As CRP was the most widely reported inflammatory biomarker, absolute change in CRP from observational and randomised studies and placebo-subtracted change in CRP from randomised controlled trials were correlated with the aforementioned variables, where available. In order to stratify participant co-morbidity, a validated multiscore co-morbidity score based on mortality by Corrao et al was used to assign a score to each study (supplementary table 4) which was then correlated with change in CRP (28). In order to correlate pharmacological intensity with change in CRP, studies including participants on no antidiabetic medications received a 0, a single agent received a 1 and more than one agent received a 2 (supplementary table 4).

Statistics

Using IBM SPSS (version 25) parametric tests, and where relevant non-parametric tests, were used to assess for linear correlations. Weighted means were weighted by study number (n). Where available, results from comparison with placebo/ standard care were preferentially included over comparison with baseline. Funnel plots used to visually assess for publication bias and tests for heterogeneity including the I^2 statistic were performed using Review Manager (RevMan) 5.3, Copenhagen: The Nordic Cochrane centre 2014. Control-subtracted change takes account of both the control and active group within a single value. This was calculated by subtracting the control group value from the SGLT2i group value for both baseline and follow-up, before subtracted the resultant baseline value from the follow-up value. Where possible standard deviation (SD) were estimated from available information using the validated calculation outlined in the Cochrane Handbook (29). A p value of <0.05 was considered statistically significant**.**

Results

From the 23 clinical studies identified, there were 1654 participants (1361 inflammation, 201 oxidative stress, 92 both) treated with SGLT2is with data relating to the relevant biomarkers. The included studies were heterogeneous in design but predominantly randomised controlled trials (15 randomised clinical trials, 8 observational studies). All of the data analysed were collected prospectively. Three publications involved post-hoc analyses of previous prospective trials:- two from the CANagliflozin Treatment And Trial Analysis versus SUlphonylurea (CANTATA-SU) trial (30, 31) and 1 from Petrykiv et al (32).

Participant characteristics can be found in table 1. All participants had T2DM, with most studies documenting a maximum HbA1c of approximately 10%. Five studies included participants who were overweight (BMI >25kg/m^2) or had the metabolic syndrome (33-37); three studies included participants with non-alcoholic liver disease (NAFLD) or non-alcoholic steatohepatitis (NASH) (36, 38, 39); one study included participants with HF (40); and one study recruited participants with coronary artery disease (41). The median number of participants recruited in the studies was 35, with a range from 11-296. All participants were aged 18 years or above, and 57.9% of participants were male. Studies were mainly composed of Japanese participants (33, 34, 37-45); but there were also participants from Germany (46), the Netherlands (32), Sweden (30), the United States of America (47), Spain (48), Italy (49), China (50), and worldwide (31, 51, 52). The daily doses of the SGLT2i was 2.5mg for luseogliflozin, 100mg-300mg for canagliflozin (mostly 100mg), 10mg-25mg for empagliflozin, and 5mg-10mg for dapagliflozin. The median duration of follow-up

was 24 weeks (range: 2 days to 104 weeks). All 6 biomarkers for inflammation and oxidative stress (CRP, adiponectin, IL6, TNF-α, 8-iso-PGF2α and 8-OHdG) showed significant heterogeneity (I^2 >75%, p < 0.10) (analysis not included).

Within the studies, five studies did not report on the loss of participants to follow-up (33-35, 46, 50). In addition, ≥5% loss to follow-up was observed in eight studies (30, 31, 36, 38, 42, 44, 48, 51); and loss to follow-up was unequal across treatment arms in four studies (30, 31, 42, 51). One study utilised a modified intention-to-treat analysis (32). Of our selected inflammatory biomarkers (CRP, adiponectin, IL6 and TNF-α) and the one most reported oxidative stress biomarker (8-iso-PGF2α), the strength of evidence as identified by the GRADE system is summarised in supplementary table 2. The quality of evidence from randomised studies was considered high for 8-iso-PGF2α, moderate for CRP, adiponectin, IL6 and TNF-α, and low for all biomarkers from observational studies. The RoB 2 tool was used to assess 12 randomised studies for risk of bias (supplementary table 3a). Six studies were thought to be of '*low*' risk of bias (32, 36, 39, 44, 45, 52); seven studies as warranting *'some concern' (30, 31, 33, 37, 41, 46, 50)* and two studies as being at *'high'* risk of bias (40, 51). The ROBINS-I tool was used to assess eight observational studies for risk of bias (supplementary table 3b). Six studies were considered '*moderate'* risk of bias (34, 35, 38, 43, 47, 49), and two studies were considered '*serious'* risk of bias (42, 48). Of the 23 studies, three did not comment on gender identity and nine studies included males with a prevalence ≥70% (33, 36, 37, 40, 41, 43, 45, 47, 48).

Inflammatory biomarkers

Our four selected inflammatory biomarkers were:- CRP in 12 studies; adiponectin in 8 studies; IL6 in 5 studies; and TNF-α in 4 studies. Results are summarised in table 2a and 2b.

C-reactive protein (CRP)

Of the 12 studies (7 randomised, 5 observational, n=732) which evaluated CRP, 83% (10/12) demonstrated a reduction in hs-CRP or CRP compared to pre-treatment levels or placebo (30, 33, 35-38, 40, 42, 48, 52). All studies show results of change in CRP compared with baseline, and 6/12 studies also reported comparison with standard care (2/12) (30, 42) or placebo (4/12) (33, 36, 46, 52). The weighted mean reduction for absolute change in CRP was -0.228 (standard deviation 0.197) and the weighted mean percentage change was 27.6% (standard deviation 30.4%). These reductions showed marked variation which might be explained by the heterogeneity in the designs of the studies. 42% (5/12) reported statistically significant reductions in CRP associated with SGLT2is (33, 35, 38, 40, 48). Of these 4 studies, the study described by Hattori, was the largest randomised study and demonstrated a marked significant decrease in hs-CRP associated with empagliflozin therapy at a dose of 10mg/day (-74.4% compared to placebo and -55.6% compared to pre-treatment levels at 12 months) (33). In the CANOSSA trial (prospective, open-label, add-on trial of canagliflozin for diabetes mellitus and stable chronic HF) (40) during the 12 months follow-up there was a significant reduction in hs-CRP (-46.2%) associated with canagliflozin therapy at a dose of 100mg/day. However, CRP (non hs-CRP) was numerically reduced, though not significantly. The smaller, observational study by Tobita et al, examined 6 months of therapy with dapagliflozin (5mg/day), and

observed a significant disease (-53.8%) in CRP compared to pre-treatment levels (38). Similarly, a randomised study by Okamoto et al, reported a -33.3% reduction in CRP at 12 weeks associated with 5mg/day of dapagliflozin (35). Iannantuoni et al, in a small (n=17), observational study which collected hs-CRP samples from older (40-70 years) participants with longstanding T2DM (>10 years) after 24 weeks of 10mg/day empagliflozin (48). Compared with baseline, hs-CRP was significantly reduced. The absence of a significant decrease in CRP in the other studies might be explained by a combination of:- (i) the recruitment of participants with fewer co-morbidities and hence a lower co-morbidity score (30, 42, 46, 52); (ii) the exclusion of participants with CRP levels above a prespecified level (e.g. >10mg/L (24); >5mg/L) (46); (iii) small participant study numbers which were insufficiently powered (n≤20) (37, 42, 43); (iv) inadequate length of follow-up (≤3 months) (36, 42, 43, 46); (v) observational study design and thus risk of bias (42, 43); and (vi) authors did not comment on statistical significance (30).

Associations with change in c-reactive protein (CRP)

Absolute change in CRP from baseline to follow-up was not significantly associated with:- (i) absolute change in HbA1c (n=8, r=-0.122, p=0.773, figure 2), (ii) participant co-morbidity (n=10, r=0.288, p=0.419), (iii) pharmacological intensity (n=10, r=0.038, p=0.918), (iv) study follow-up period (n=10, r=0.263, p=0.463), (v) SGLT2i concentration (n=10, r=0.116, p=0.749), (vi) absolute change in eGFR (n=6, r=-0.545, p=0.264), (vii) absolute change in BMI (n=4, r=-0.077, p=0.923) and (viii) absolute change in HOMA-IR. Looking specifically at randomised controlled trials, placebo-subtracted change in CRP was not significantly associated with:- (i)

placebo-subtracted change in HbA1c (n=4, r=0.400, p=0.600), (ii) study length (n=4, -0.641, p=0.359), (iii) SGLT2i concentration (n=4, r=0.177, p=0.823), (iv) participant co-morbidity (n=5, r=0.129, p=0.836) and (v) pharmacological intensity (n=5, r=0.198, p=0.750).

Adiponectin

With respect to adiponectin, eight studies (5 randomised, 3 observational, n=553) investigated changes in adiponectin following SGLT2is. All studies report data on change in adiponectin relative to baseline, and 4/8 report comparison with either standard care (2/8) (30, 39) or placebo (2/8) (36, 51). Of these eight studies, 75% (6/8) described an increase in adiponectin, (30, 34, 35, 38, 39, 51) and of which four studies (4/8) described were significant (34, 35, 38, 39) and two studies did not comment on the statistical significance. The largest randomised study (n=282) did not comment on significance but showed an increase in adiponectin (51). Bailey et al, described an increase in adiponectin associated with dapagliflozin with 1, 2.5 and 5mg/day doses at 3 months compared to placebo and from pre-treatment levels in treatment naïve participants with T2DM. Similarly, Aso et al, report a significant increase (+50.0%) in adiponectin following 3 months of therapy with dapagliflozin (5mg/day) compared to pre-treatment levels and a control-subtracted increase of +56.0% (39). Garvey et al, described a non-significant increase in adiponectin (+17.1%) following 52 weeks of canagliflozin 300mg/day compared with glimepiride 6mg or 8mg/day in a sample of participants with fewer co-morbidities (30). Other studies, which showed a significant increase in adiponectin, were generally smaller and showed increases of +22.9% (dapagliflozin 5mg/day, n=11, duration 6 months)

(34); +9.4% (canagliflozin 100mg/day, n=15, duration 7 days) (34); and +33.3% (dapagliflozin 5mg/day, n=27, duration of 3 months) (35). A further three studies investigating dapagliflozin and luseogliflozin described small, non-significant decreases in adiponectin (32, 36, 37).

Interleukin 6 (IL6)

There were four published datasets (3 randomised, 1 observational, n=347, from 5 studies) which examined changes in IL6 following use of SGLT2i (30-32, 37, 47). 4/5 studies reported data on IL6 compared with baseline, (30, 32, 37, 47) and 3/5 reported data compared with standard care (2/5) (30, 31) or placebo (1/5) (32). Both Heerspink and Garvey et al, performed a post-hoc analysis from the CANTATA-SU trial (30, 31). Of the studies that investigated changes in IL6 compared to pre-treatment levels, placebo or standard care, all showed a decrease in plasma IL6, with 80% (4/5) of these describing significant decreases (31, 32, 47, 48). Heerspink et al, measured plasma IL6 in 296 samples from the CANTATA-SU trial (31). They observed a significant and substantial decrease in IL6 associated with 100mg/day and 300mg/day of canagliflozin following 104 weeks of therapy (-26.3% and -26.6%, respectively). Tan and Tan, in a study comparing the efficacy of empagliflozin with canagliflozin observed that following 6 months of treatment, empagliflozin 10mg/day was superior and was associated with a significant decrease in IL6 by -52.0% (47). The randomised study by Dekkers et al, observed a significant decrease in IL6 compared with baseline following dapagliflozin therapy at a dose of 10mg/day (32). Bouchi et al, reported a non-significant decrease in IL6 (-33.0%) with luseogliflozin

(2.5mg/day) compared with baseline, but was not powered for the purpose of examining change in IL6, including only 19 participants (37).

Tumour necrosis factor alpha (TNF-α)

Four studies (2 randomised, 2 observational, n=187) have investigated changes in TNF-α (30, 34, 41, 47), all of which reported changes in TNF-α compared with baseline and 2/4 (30, 41) report changes compared with standard care. 75% (3/4) described a decrease (34, 41, 47) with 50% (2/4) showing a statistically significant decrease with SGLT2is (41, 47). Both studies were of a moderate size with a follow-up period of 6 months. Tan and Tan (n=32), observed a significant decrease in TNF-α of -37.4% (47) following treatment with 10mg/day of empagliflozin in a sample of males. Sato et al (n=40), observed a decrease of -22.1% associated with dapagliflozin (dose unspecified) in a sample of T2DM participants with coronary artery disease (41). The remaining two studies did not show significant reductions, but of note, these recruited participants with fewer co-morbidities, (30) and were insufficiently powered with a short follow-up (34).

Oxidative Stress

Our two selected oxidative stress biomarkers were:- 8-iso-PGF2α in 4 studies and 8-OHdG in 2 studies. Results are summarised in table 2a and 2b.

8-iso-prostaglandin F2α (8-iso-PGF2α)

Four studies (3 randomised, 1 observational, n=146) investigated 8-iso-PGF2α levels with three reporting urinary levels (36, 45, 49) and one reporting serum levels (50). 75% of these studies report significant decreases in 8-iso-PGF2α levels following use of SGLT2is. The randomised trial by Nishimura et al, (n=60) observed a significant decrease in fasting urinary 8-iso-PGF2α at 28 days following treatment with empagliflozin 10mg/day (-45.5%) and empagliflozin 25mg/day (-50.5%) relative to placebo (45). Similarly the observational study by Solini et al, (n=16) reported a significant decrease (-30.3%) in urinary 8-iso-PGF2α levels 2 days after a single 10mg dose of dapagliflozin (49). Similar serum measurements have been observed in the randomised trial (n=28) by Zhou et al, in which 24 weeks of dapagliflozin (between 5mg-10mg) was associated with a significant decrease in serum 8-iso-PGF2α levels compared with placebo (50). Eriksson et al, studied T2DM patients with concomitant NAFLD and reported non-significant increases in urinary 8-iso-PGF2α/creatinine ratio (+12.3%) and urinary 2,3-dinor-8-iso-PGF2α/creatinine (+18.2%) following 12 weeks of 10mg/day dapaglifozin compared with placebo (36). At 24 hours post-drug administration, they also observed a significant decrease in urinary 8-iso-PGF2α following empagliflozin 25mg/day (-43.1%) and a non-significant decrease following empagliflozin 10mg/day (-24.7%).

8-hydroxy-2'-deoxyguanosine (8-OHdG)

Two studies (1 randomised, 1 observational, n=95) report urinary 8-OHdG levels (34, 44) and both report a significant decrease following SGLT2is. Shigiyama et al, published the DEFENCE (Dapagliflozin EFfectiveness on vascular ENdothelial function and glycemic Control in patients with Early-stage type 2 diabetes mellitus) study (n=80), which examined participants receiving 750mg/day of metformin and

who were randomised to dapagliflozin 5mg/day dapagliflozin (SGLT2i) or metformin 1500mg/day (control) (44). At 16 week follow-up, they observed a significant decrease (-34.8%) in the urinary 8-OHdG/creatinine ratio in the dapagliflozin group compared to control. Of note, this change was non-significant compared to baseline (-13.0%). The observational study in stable, insulin-treated patients with T2DM by Matsumura et al (n=15), reported a significant decrease (-5.3%) in urinary 8-OHdG/creatinine ratio relative to pre-treatment levels following 3 days of canagliflozin 100mg/day (34).

Discussion

This systematic review of 23 studies with 1654 patients summarises the current available literature, which examine the effects of SGLT2is on inflammatory and oxidative stress biomarkers. We have observed that SGLT2is were associated with significant decreases in CRP, IL6, TNF-α, 8-iso-PGF2α, and 8-OhdG, along with significant increases in adiponectin. The association of T2DM with inflammation and oxidative stress is well established (53-55). Indeed, many diabetes medications have anti-inflammatory and anti-oxidant effects (56, 57). Our data support the hypothesis that SGLT2is decrease inflammation and oxidative stress in T2DM.

Beyond the markers examined in this review, other less investigated biomarkers such as leptin also show SGLT2i-associated decreases (47). Interestingly inflammatory markers specific to atherosclerosis, such as RLP-C, are limited but demonstrate significant reductions (33, 40). By clarifying the effect of SGLT2is on inflammatory and oxidative stress pathways this could contribute to establishing new therapeutics for patients with and without diabetes

Association between clinical and laboratory-based evidence

There is consistent evidence from animal models that SGLT2is ameliorate the inflammatory and oxidative stress profile observed with T2DM. Tahara et al, used a murine model of T2DM to demonstrate significant reductions in plasma CRP, TNF-α and IL6 following 4 weeks of therapy with 3mg/kg/day of ipragliflozin (58). In addition, other published studies report reductions in IL6 and TNF-α with SGLT2is. Studies using cell culture cytotoxic assays in human endothelial cells (59), human and mice immune cells (60) have observed that canagliflozin is associated with a reduction in IL6 and TNF-α. Of interest, no reduction has been observed in these cytokines in endothelial cells treated empagliflozin or dapagliflozin (59). Treatment with empagliflozin has been associated with a decreased expression of IL6 in a murine model of DKD (61), along with TNF-α in high-fat-diet induced obese mice (62). Gene expression microarrays of mouse adipose tissue have shown that an obese murine model of T2DM is associated with down-regulation of adipocytokines including adiponectin, and this is re-upregulated by dapagliflozin (63). In the mouse model of atherosclerosis (apolipoprotein E knockout), canagliflozin was associated with reduced expression of inflammatory molecules including monocyte chemoattractant protein-1 (MCP-1) and VCAM-1 (64). The authors suggest that this was associated with histological changes demonstrating a slowing of atherosclerosis. Within these studies, it is difficult to compare study design, follow-up time and dose as these experiments are based in animal models.

Very few animal model studies have examined markers of iso-8-PGF2α. Salim et al, observed reductions in 8-OHdG in a murine model of T2DM following treatment with

3mg/kg/day of ipragliflozin (65). In addition Osorio el al, described an increase in catalase and a decrease in glutathione peroxidase in the diabetic rat kidney cortex and medulla following treatment with phlorizin (66). Using streptozotocin toxicity as a model of diabetes, Oelze et al, reported that NADPH oxidase activity in rat heart membranes was increased following streptozotocin treatment, but dose-dependently inhibited by SGLT2is (67). Such changes are consistent with the findings of our systematic review.

How do SGLT2 inhibitors exert such effects?

The mechanism by which SGLT2is decrease inflammation in T2DM remains unclear. There appears to be an interplay between oxidative stress and inflammation that is independent of reduced inflammation as a result of improved glycaemic control (68). One proposed hypothesis suggests that SGLT2is suppress the up-regulation of the NOD-like receptor family pyrin domain containing 3 (NLRP3) inflammasome (69). Another hypothesis relates to a possible role that SGLT2is inhibit intracellular glucose metabolism leading to increased autophagy and a resultant dampening of the inflammatory response (60).

Limitations of study

The current evidence base consists of a heterogeneous group of studies, predominantly at '*moderate'* risk of bias, and often underpowered to reliably detect changes in serum inflammatory and oxidative stress biomarkers. Due to study design heterogeneity and unavailability of data, it was not possible to perform a meta-analysis. A focus of further studies should be to increase the power of any results in order to enable a more accurate interpretation of findings. 39% of included

studies included ≥70% male participants, although this only amounted to 57.9% of total participants being male in this study. Moreover, the prevalence of T2DM is higher in males (70) and therefore the generalisability of our results is unlikely to be compromised. Another limitation of our study was the selection of adequate biomarkers, for instance, uric acid was excluded from our study as it has both anti-oxidant and pro-oxidant properties depending on its location, in plasma or cytoplasm (71). There is also emerging evidence that certain markers believed to be gold-standard are imperfect markers of inflammation or oxidative stress (72). Future clinical studies must attempt to ensure the validity of any biomarkers investigated.

Conclusion

Despite the heterogeneity of the available published studies and based on predominantly '*moderate'* quality data, the findings of this review supports evidence that SGLT2is reduce inflammation and oxidative stress associated with T2DM at short- and long-term follow-up. This may partly explain the CV and renal benefits of SGLT2is. The beneficial effect of SGLT2is on measurements of serum CRP appear to be independent of many study design and clinically relevant variables, including HbA1c.

Acknowledgements

Jonathan J. H. Bray and Harri Foster-Davies would like to acknowledge Mr. Rhys Whelan for his support in assisting in conducting the evidence search.

Conflicts of Interest

The authors declare no conflicts of interest in this review.

Contribution statement

JJHB is responsible for writing the initial manuscript, design, data collection and analysis. HFD made significant contributions to writing of the manuscript, data collection and analysis. JWS conceptualised and supervised the project, and thoroughly reviewed design, data collection, analysis and the writing of the final manuscript.

References

1. Einarson TR, Acs A, Ludwig C, Panton UH. Prevalence of cardiovascular disease in type 2 diabetes: a systematic literature review of scientific evidence from across the world in 2007-2017. Cardiovascular Diabetology. 2018;17:19.
2. US Department of Health and Human Services CfDCaP. Centers for Disease Control and Prevention. National Diabetes Fact Sheet: General Information and National Estimates on Diabetes in the United States, 2014. https://nccd.cdc.gov/CKD/Data.aspx?CategoryID=1#12014. Accessed November 15, 2019.
3. Adler AI, Stevens RJ, Manley SE, Bilous RW, Cull CA, Holman RR, et al. Development and progression of nephropathy in type 2 diabetes: The United Kingdom Prospective Diabetes Study (UKPDS 64). Kidney International. 2003;63(1):225-32.
4. Sarwar N, Gao P, Seshasai SRK, Gobin R, Kaptoge S, Di Angelantonio E, et al. Diabetes mellitus, fasting blood glucose concentration, and risk of vascular disease: a collaborative meta-analysis of 102 prospective studies. Lancet. 2010;375(9733):2215-22.
5. Kengne AP, Turnbull F, MacMahon S. The Framingham Study, diabetes mellitus and cardiovascular disease: turning back the clock. Prog Cardiovasc Dis. 2010;53(1):45-51.
6. Zinman B, Wanner C, Lachin JM, Fitchett D, Bluhmki E, Hantel S, et al. Empagliflozin, Cardiovascular Outcomes, and Mortality in Type 2 Diabetes. New England Journal of Medicine. 2015;373(22):2117-28.
7. Neal B, Perkovic V, Mahaffey KW, de Zeeuw D, Fulcher G, Erondu N, et al. Canagliflozin and Cardiovascular and Renal Events in Type 2 Diabetes. New England Journal of Medicine. 2017;377(7):644-57.

8. Wiviott SD, Raz I, Bonaca MP, Mosenzon O, Kato ET, Cahn A, et al. Dapagliflozin and Cardiovascular Outcomes in Type 2 Diabetes. New England Journal of Medicine. 2019;380(4):347-57.
9. Neuen BL, Ohkuma T, Neal B, Matthews DR, de Zeeuw D, Mahaffey KW, et al. Cardiovascular and Renal Outcomes With Canagliflozin According to Baseline Kidney Function: Data From the CANVAS Program. Circulation. 2018;138(15):1537-50.
10. Vasilakou D, Karagiannis T, Athanasiadou E, Mainou M, Liakos A, Bekiari E, et al. Sodium-Glucose Cotransporter 2 Inhibitors for Type 2 Diabetes A Systematic Review and Meta-analysis. Annals of Internal Medicine. 2013;159(4):262-+.
11. Perkovic V, Jardine MJ, Neal B, Bompoint S, Heerspink HJL, Charytan DM, et al. Canagliflozin and Renal Outcomes in Type 2 Diabetes and Nephropathy. New England Journal of Medicine. 2019;380(24):2295-306.
12. Toyama T, Neuen BL, Jun M, Ohkuma T, Neal B, Jardine MJ, et al. Effect of SGLT2 inhibitors on cardiovascular, renal and safety outcomes in patients with type 2 diabetes mellitus and chronic kidney disease: A systematic review and meta-analysis. Diabetes Obesity & Metabolism. 2019;21(5):1237-50.
13. Zelniker TA, Wiviott SD, Raz I, Im K, Goodrich EL, Bonaca MP, et al. SGLT2 inhibitors for primary and secondary prevention of cardiovascular and renal outcomes in type 2 diabetes: a systematic review and meta-analysis of cardiovascular outcome trials. Lancet. 2019;393(10166):31-9.
14. Davies MJ, D'Alessio DA, Fradkin J, Kernan WN, Mathieu C, Mingrone G, et al. Management of Hyperglycemia in Type 2 Diabetes, 2018. A Consensus Report by the American Diabetes Association (ADA) and the European Association for the Study of Diabetes (EASD). Diabetes Care. 2018;41(12):2669-701.
15. Bonora BM, Avogaro A, Fadini GP. Extraglycemic Effects of SGLT2 Inhibitors: A Review of the Evidence. Diabetes Metabolic Syndrome and Obesity-Targets and Therapy. 2020;13:161-74.
16. Donath MY. Targeting inflammation in the treatment of type 2 diabetes: time to start. Nature Reviews Drug Discovery. 2014;13(6):465-76.
17. Folli F, Corradi D, Fanti P, Davalli A, Paez A, Giaccari A, et al. The role of oxidative stress in the pathogenesis of type 2 diabetes mellitus micro- and macrovascular complications: avenues for a mechanistic-based therapeutic approach. Curr Diabetes Rev. 2011;7(5):313-24.
18. Libby P. Inflammation in Atherosclerosis. Arteriosclerosis Thrombosis and Vascular Biology. 2012;32(9):2045-51.
19. Vallon V, Komers R. Pathophysiology of the Diabetic Kidney. Comprehensive Physiology. 2011;1(3):1175-232.
20. Ridker PM, Everett BM, Thuren T, MacFadyen JG, Chang WH, Ballantyne C, et al. Antiinflammatory Therapy with Canakinumab for Atherosclerotic Disease. New England Journal of Medicine. 2017;377(12):1119-31.
21. Vallon V. The Mechanisms and Therapeutic Potential of SGLT2 Inhibitors in Diabetes Mellitus. In: Caskey CT, editor. Annual Review of Medicine, Vol 66. Annual Review of Medicine. 66. Palo Alto: Annual Reviews; 2015. p. 255-70.
22. Han J, Oh T, Lee G, Maeng H, Lee D, Kim K, et al. The beneficial effects of empagliflozin, an SGLT2 inhibitor, on atherosclerosis in ApoE (-/-) mice fed a western diet. Diabetologia. 2017;60(2):364-76.
23. Yaribeygi H, Atkin SL, Butler AE, Sahebkar A. Sodium-glucose cotransporter inhibitors and oxidative stress: An update. Journal of Cellular Physiology. 2019;234(4):3231-7.
24. Tahara A, Takasu T. Effects of the SGLT2 inhibitor ipragliflozin on various diabetic symptoms and progression of overt nephropathy in type 2 diabetic mice. Naunyn-Schmiedebergs Archives of Pharmacology. 2018;391(4):395-406.

25. Guyatt GH, Oxman AD, Vist GE, Kunz R, Falck-Ytter Y, Alonso-Coello P, et al. GRADE: an emerging consensus on rating quality of evidence and strength of recommendations. British Medical Journal. 2008;336(7650):924-6.
26. Sterne JAC, Savovic J, Page MJ, Elbers RG, Blencowe NS, Boutron I, et al. RoB 2: a revised tool for assessing risk of bias in randomised trials. Bmj-British Medical Journal. 2019;366:8.
27. Sterne JAC, Hernan MA, Reeves BC, Savovic J, Berkman ND, Viswanathan M, et al. ROBINS-I: a tool for assessing risk of bias in non-randomised studies of interventions. Bmj-British Medical Journal. 2016;355:7.
28. Corrao G, Rea F, Di Martino M, De Palma R, Scondotto S, Fusco D, et al. Developing and validating a novel multisource comorbidity score from administrative data: a large population-based cohort study from Italy. Bmj Open. 2017;7(12):8.
29. Higgins JPT, Green S (editors). Cochrane Handbook for Systematic Reviews of Interventions Version 5.1.0 [updated March 2011]. The Cochrane Collaboration, 2011. Available from https://handbook-5-1.cochrane.org/chapter_16/16_1_3_2_imputing_standard_deviations_for_changes_from_baseline.htm.
30. Garvey WT, Van Gaal L, Leiter LA, Vijapurkar U, List J, Cuddihy R, et al. Effects of canagliflozin versus glimepiride on adipokines and inflammatory biomarkers in type 2 diabetes. Metabolism: Clinical & Experimental. 2018;85:32-7.
31. Heerspink HJL, Perco P, Mulder S, Leierer J, Hansen MK, Heinzel A, et al. Canagliflozin reduces inflammation and fibrosis biomarkers: a potential mechanism of action for beneficial effects of SGLT2 inhibitors in diabetic kidney disease. Diabetologia. 2019;62(7):1154-66.
32. Dekkers CCJ, Petrykiv S, Laverman GD, Cherney DZ, Gansevoort RT, Heerspink HJL. Effects of the SGLT-2 inhibitor dapagliflozin on glomerular and tubular injury markers. Diabetes Obes Metab. 2018;20(8):1988-93.
33. Hattori S. Anti-inflammatory effects of empagliflozin in patients with type 2 diabetes and insulin resistance. Diabetology & Metabolic Syndrome. 2018;10:7.
34. Matsumura M, Nakatani Y, Tanka S, Aoki C, Sagara M, Yanagi K, et al. Efficacy of Additional Canagliflozin Administration to Type 2 Diabetes Patients Receiving Insulin Therapy: Examination of Diurnal Glycemic Patterns Using Continuous Glucose Monitoring (CGM). Diabetes Therapy. 2017;8(4):821-7.
35. Okamoto A, Yokokawa H, Sanada H, Naito T. Changes in Levels of Biomarkers Associated with Adipocyte Function and Insulin and Glucagon Kinetics During Treatment with Dapagliflozin Among Obese Type 2 Diabetes Mellitus Patients. Drugs in R&D. 2016;16(3):255-61.
36. Eriksson JW, Lundkvist P, Jansson PA, Johansson L, Kvarnstrom M, Moris L, et al. Effects of dapagliflozin and n-3 carboxylic acids on non-alcoholic fatty liver disease in people with type 2 diabetes: a double-blind randomised placebo-controlled study. Diabetologia. 2018;61(9):1923-34.
37. Bouchi R, Terashima M, Sasahara Y, Asakawa M, Fukuda T, Takeuchi T, et al. Luseogliflozin reduces epicardial fat accumulation in patients with type 2 diabetes: a pilot study. Cardiovascular Diabetology. 2017;16:9.
38. Tobita H, Sato S, Miyake T, Ishihara S, Kinoshita Y. Effects of Dapagliflozin on Body Composition and Liver Tests in Patients with Nonalcoholic Steatohepatitis Associated with Type 2 Diabetes Mellitus: A Prospective, Open-label, Uncontrolled Study. Current Therapeutic Research-Clinical and Experimental. 2017;87:13-9.
39. Aso Y, Kato K, Sakurai S, Kishi H, Shimizu M, Jojima T, et al. Impact of dapagliflozin, an SGLT2 inhibitor, on serum levels of soluble dipeptidyl peptidase-4 in patients with type 2 diabetes and non-alcoholic fatty liver disease. International Journal of Clinical Practice. 2019;73(5):e13335.
40. Sezai A, Sekino H, Unosawa S, Taoka M, Osaka S, Tanaka M. Canagliflozin for Japanese patients with chronic heart failure and type II diabetes. Cardiovascular Diabetology. 2019;18:13.

41. Sato T, Aizawa Y, Yuasa S, Kishi S, Fuse K, Fujita S, et al. The effect of dapagliflozin treatment on epicardial adipose tissue volume. Cardiovascular Diabetology. 2018;17:9.
42. Noda T, Ebihara E, Ueno H, Sadohara K, Tanaka Y, Nagatomo Y, et al. Concurrent Use of Teneligliptin and Canagliflozin Improves Glycemic Control with Beneficial Effects on Plasma Glucagon and Glucagon-Like Peptide-1: A Single-Arm Study. Diabetes Therapy. 2019;10(5):1835-46.
43. Osonoi T, Gouda M, Kubo M, Arakawa K, Hashimoto T, Abe M. Effect of Canagliflozin on Urinary Albumin Excretion in Japanese Patients with Type 2 Diabetes Mellitus and Microalbuminuria: A Pilot Study. Diabetes Technol Ther. 2018;20(10):681-8.
44. Shigiyama F, Kumashiro N, Miyagi M, Ikehara K, Kanda E, Uchino H, et al. Effectiveness of dapagliflozin on vascular endothelial function and glycemic control in patients with early-stage type 2 diabetes mellitus: DEFENCE study. Cardiovascular Diabetology. 2017;16:12.
45. Nishimura R, Tanaka Y, Koiwai K, Inoue K, Hach T, Salsali A, et al. Effect of empagliflozin monotherapy on postprandial glucose and 24-hour glucose variability in Japanese patients with type 2 diabetes mellitus: a randomized, double-blind, placebo-controlled, 4-week study. Cardiovascular Diabetology. 2015;14:13.
46. Bosch A, Ott C, Jung S, Striepe K, Karg MV, Kannenkeril D, et al. How does empagliflozin improve arterial stiffness in patients with type 2 diabetes mellitus? Sub analysis of a clinical trial. Cardiovasc Diabetol. 2019;18(1):44.
47. Tan SA, Tan L. Empagliflozin and canagliflozin attenuate inflammatory cytokines interferon-gamma, tumor necrosis factor-alpha, interleukin-6: possible mechanism of decreasing cardiovascular risk in diabetes mellitus. Journal of the American College of Cardiology. 2018;71(11):1830-.
48. Iannantuoni F, de Maranon AM, Diaz-Morales N, Falcon R, Banuls C, Abad-Jimenez Z, et al. The SGLT2 Inhibitor Empagliflozin Ameliorates the Inflammatory Profile in Type 2 Diabetic Patients and Promotes an Antioxidant Response in Leukocytes. Journal of Clinical Medicine. 2019;8(11):11.
49. Solini A, Giannini L, Seghieri M, Vitolo E, Taddei S, Ghiadoni L, et al. Dapagliflozin acutely improves endothelial dysfunction, reduces aortic stiffness and renal resistive index in type 2 diabetic patients: a pilot study. Cardiovascular Diabetology. 2017;16:9.
50. Zhou PH, Ma JH, Li FF. Influence of SGLT2 inhibitors on plasma glycemic fluctuations and oxidative stress in patients with newly diagnosed type 2 diabetes mellitus. Diabetes-Metabolism Research and Reviews. 2016;32(SUPP 2):40-.
51. Bailey CJ, Iqbal N, T'Joen C, List JF. Dapagliflozin monotherapy in drug-naive patients with diabetes: a randomized-controlled trial of low-dose range. Diabetes Obesity & Metabolism. 2012;14(10):951-9.
52. Ferrannini E, Ramos SJ, Salsali A, Tang WH, List JF. Dapagliflozin Monotherapy in Type 2 Diabetic Patients With Inadequate Glycemic Control by Diet and Exercise A randomized, double-blind, placebo-controlled, phase 3 trial. Diabetes Care. 2010;33(10):2217-24.
53. Marques-Vidal P, Schmid R, Bochud M, Bastardot F, von Kanel R, Paccaud F, et al. Adipocytokines, Hepatic and Inflammatory Biomarkers and Incidence of Type 2 Diabetes. The CoLaus Study. Plos One. 2012;7(12):8.
54. Bertoni AG, Burke GL, Owusu JA, Carnethon MR, Vaidya D, Barr RG, et al. Inflammation and the Incidence of Type 2 Diabetes The Multi-Ethnic Study of Atherosclerosis (MESA). Diabetes Care. 2010;33(4):804-10.
55. 55. Evans JL, Goldfine ID, Maddux BA, Grodsky GM. Oxidative stress and stress-activated signaling pathways: A unifying hypothesis of type 2 diabetes. Endocrine Reviews. 2002;23(5):599-622.

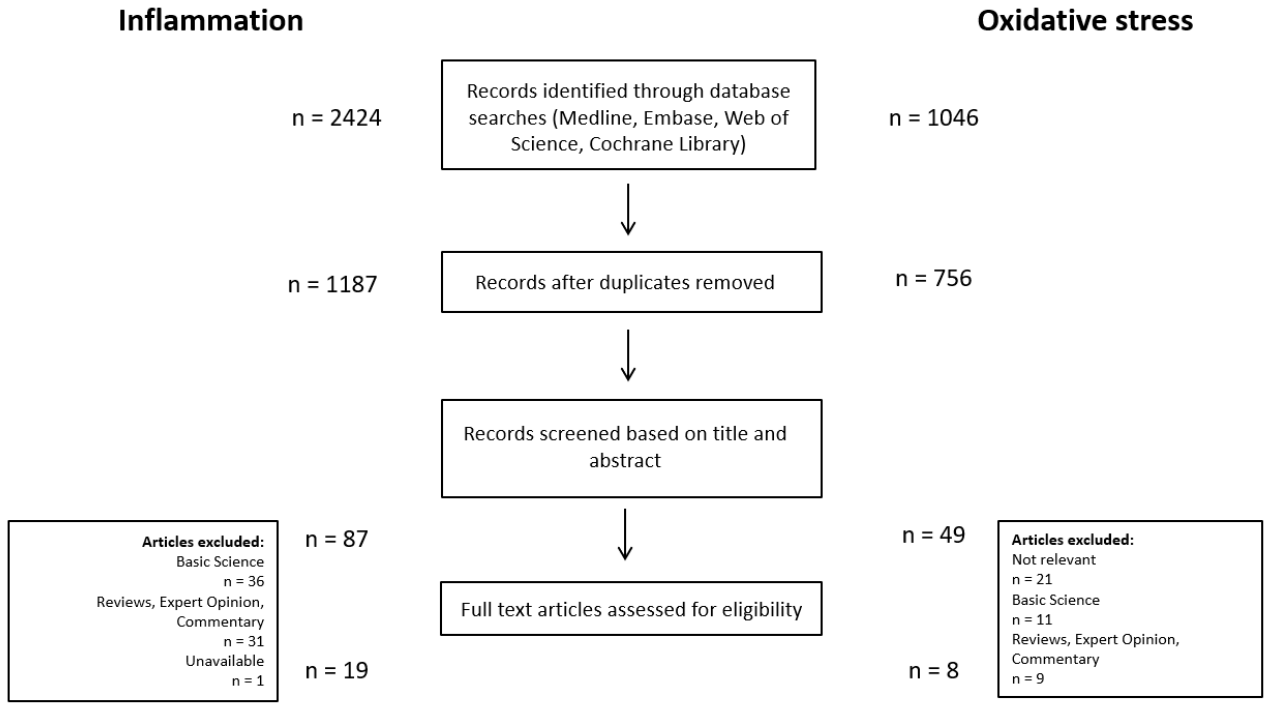
56. Yaribeygi H, Butler AE, Barreto GE, Sahebkar A. Antioxidative potential of antidiabetic agents: A possible protective mechanism against vascular complications in diabetic patients. Journal of Cellular Physiology. 2019;234(3):2436-46.
57. Kothari V, Galdo JA, Mathews ST. Hypoglycemic agents and potential anti-inflammatory activity. Journal of Inflammation Research. 2016;9:27-38.
58. Tahara A, Kurosaki E, Yokono M, Yamajuku D, Kihara R, Hayashizaki Y, et al. Effects of SGLT2 selective inhibitor ipragliflozin on hyperglycemia, hyperlipidemia, hepatic steatosis, oxidative stress, inflammation, and obesity in type 2 diabetic mice. European Journal of Pharmacology. 2013;715(1-3):246-55.
59. Mancini SJ, Boyd D, Katwan OJ, Strembitska A, Almabrouk TA, Kennedy S, et al. Canagliflozin inhibits interleukin-1 beta-stimulated cytokine and chemokine secretion in vascular endothelial cells by AMP-activated protein kinase-dependent and -independent mechanisms. Scientific Reports. 2018;8:14.
60. Xu CK, Wang W, Zhong J, Lei F, Xu NH, Zhang YO, et al. Canagliflozin exerts anti-inflammatory effects by inhibiting intracellular glucose metabolism and promoting autophagy in immune cells. Biochemical Pharmacology. 2018;152:45-59.
61. Gembardt F, Bartaun C, Jarzebska N, Mayoux E, Todorov VT, Hohenstein B, et al. The SGLT2 inhibitor empagliflozin ameliorates early features of diabetic nephropathy in BTBR ob/ob type 2 diabetic mice with and without hypertension. American Journal of Physiology-Renal Physiology. 2014;307(3):F317-F25.
62. Xu L, Nagata N, Nagashimada M, Fen ZG, Ni YH, Chen GL, et al. SGLT2 Inhibition by Empagliflozin Promotes Fat Utilization and Browning and Attenuates Inflammation and Insulin Resistance by Polarizing M2 Macrophages in Diet-induced Obese Mice. Ebiomedicine. 2017;20:137-49.
63. Nishitani S, Fukuhara A, Shin J, Okuno Y, Otsuki M, Shimomura I. Metabolomic and microarray analyses of adipose tissue of dapagliflozin-treated mice, and effects of 3-hydroxybutyrate on induction of adiponectin in adipocytes. Scientific Reports. 2018;8:11.
64. Nasiri-Ansari N, Dimitriadis GK, Agrogiannis G, Perrea D, Kostakis ID, Kaltsas G, et al. Canagliflozin attenuates the progression of atherosclerosis and inflammation process in APOE knockout mice. Cardiovascular Diabetology. 2018;17:12.
65. Salim HM, Fukuda D, Yagi S, Soeki T, Shimabukuro M, Sata M. Glycemic Control with Ipragliflozin, a Novel Selective SGLT2 Inhibitor, Ameliorated Endothelial Dysfunction in Streptozotocin-Induced Diabetic Mouse. Front Cardiovasc Med. 2016;3:43.
66. Osorio H, Coronel I, Arellano A, Pacheco U, Bautista R, Franco M, et al. Sodium-Glucose Cotransporter Inhibition Prevents Oxidative Stress in the Kidney of Diabetic Rats. Oxidative Medicine and Cellular Longevity. 2012:7.
67. Welschof P, Oelze M, Kroller-Schon S, Jansen T, Hausding M, Mikhed Y, et al. The sodium-glucose cotransporter 2 inhibitor empagliflozin improves diabetic complications in the streptozotocin type 1 diabetes mellitus model by interfering with glucotoxicity and rescue of beta-cell function. Arteriosclerosis, Thrombosis, and Vascular Biology Conference: American Heart Association's Arteriosclerosis, Thrombosis and Vascular Biology. 2014;34(SUPPL. 1).
68. Wang SF, Li YB, Fan JJ, Zhang XY, Luan JY, Bian Q, et al. Interleukin-22 ameliorated renal injury and fibrosis in diabetic nephropathy through inhibition of NLRP3 inflammasome activation. Cell Death & Disease. 2017;8:10.
69. Sharma A, Tate M, Mathew G, Vince JE, Ritchie RH, de Haan JB. Oxidative Stress and NLRP3-Inflammasome Activity as Significant Drivers of Diabetic Cardiovascular Complications: Therapeutic Implications. Frontiers in Physiology. 2018;9:15.
70. UK D. Middle-aged men twice as likely to have diabetes as women https://www.diabetes.org.uk/about_us/news_landing_page/middle-aged-men-twice-as-likely-to-have-diabetes-as-women2009. Accessed January 28, 2020.

71. Verma S. Potential Mechanisms of Sodium-Glucose Co-Transporter 2 Inhibitor-Related Cardiovascular Benefits. American Journal of Medicine. 2019;132(10):S39-S48.
72. Van't Erve TJ, Lih FB, Kadiiska MB, Deterding LJ, Eling TE, Mason RP. Reinterpreting the best biomarker of oxidative stress: The 8-iso-PGF(2 alpha)/PGF(2 alpha), ratio distinguishes chemical from enzymatic lipid peroxidation. Free Radical Biology and Medicine. 2015;83:245-51.

Figure 1a: Search Strategies

No	Searched Terms - Inflammation	Searched Terms - Oxidative Stress
1	gliflozin*	gliflozin*
2	Sglt$2 inhibitor*	Sglt$2 inhibitor*
3	sglt2 inhibitor*	sglt2 inhibitor*
4	sodium glucose cotransporter 2 inhibitor*	sodium glucose cotransporter 2 inhibitor*
5	OR/1-4	OR/1-4
6	Inflammation	Oxidative Stress
7	Biomarker*	Biomarker*
8	6 OR 7	6 OR 7
9	5 AND 8	5 AND 8

Figure 1b: PRISMA Flow Diagram



Legend

742 (Figure 1a) Search strategy used to search Medline, Embase, Web of Science, and the
743 Cochrane Library. (Figure 2b) Resulting articles found and reasons for exclusion.
744 Keywords: (*) = ending variant, ($) = truncation.

Table 1: Study and population characteristics

Study	Type of Study	Number randomised (n)	Mean age (years ±SD)	Sex split (% male)	Loss to follow-up (%)	Inclusion criteria
Iannantuoni et al, 2019 (48)	Prospective, open-label observational	17	60.8 (±10.2)	73%	11.8%	T2DM
Bosch et al, 2019 (46)	Prospective, double-blind, randomised, placebo-controlled trial	58	62.0 (±7.0)	59%	-	T2DM
Heerspink et al, 2019 (31) and Garvey et al, 2018 (30)	Prospective, double-blind, randomised controlled trial (CANTATA-SU trial)	296	Heerspink: 56.3 (±9.1) Garvey: 58 (±8.8)	51%	19.9% Equal	T2DM
Aso et al, 2019 (39)	Prospective, open-label, randomised, blinded endpoint trial	57	57.1 (±12.5)	-	4.8% Equal	T2DM, NAFLD
Sezai et al, 2019 (40)	Prospective, open-label, randomised controlled trial (CANOSSA trial)	35	71.4 (±11.3)	78%	0%	T2DM, HF
Noda et al, 2019 (42)	Observational, open-label	12	61.3 (±2.5)	25%	7.7% Unequal	T2DM
Dekkers et al, 2018 (32)	Prospective, double-blind, cross-over randomised placebo-controlled trial	31	62.0 (±8.1)	77.4%	0 % modified intention-to-treat	T2DM
Eriksson et al, 2018 (36)	Multicentre, double-blind, prospective, randomised placebo-controlled double-dummy four-armed parallel-group trial (EFFECT-II trial)	42	65.3 (±6.3)	79%	9.5% Equal	T2DM, NAFLD, BMI (25–40 kg/m^2)
Hattori, 2018 (33)	Prospective, open-	102	57.8	77%	-	T2DM, proven insulin resistance

	label, randomised		(±11.0)			
Osonoi et al, 2018 (43)	Observational, open-label	20	62.9 (±8.6)	75%	0%	T2DM, moderate albuminuria
Tan and Tan, 2018 (47)	Conference abstract	32	52.2	100%	0%	Male, T2DM
Sato et al, 2018 (41)	Prospective, randomised controlled trial	40	67.0 (±5.0)	75%	0%	T2DM
Bouchi et al, 2017 (37)	Prospective, randomised, single-arm pilot	19	55.0 (±12.0)	74%	0%	T2DM, BMI ≥25 kg/m^2
Tobita et al, 2017 (38)	Prospective, open-label, observational, pilot	11	57.8 (±12.0)	54.5%	31.3% Single-arm	T2DM, NASH, One to three of: metabolic syndrome, hypertension and dyslipidaemia
Matsumura et al, 2017 (34)	Observational, open label, single-arm	15	52.9 (±14.4)	66.7%	-	T2DM
Solini et al. 2017 (49)	Prospective, observational	16	57 .0 (±9.0)	69%	0%	T2DM, BMI <40 kg/m^2
Shigiyama et al. 2017 (44)	Prospective, open-label, blinded-endpoint, randomised (DEFENCE study)	80	58.7 (±9.2)	64%	7.5% Equal	T2DM
Okamoto et al, 2016 (35)	Prospective, observational, open-label, single-arm	27	49.7 (±9.0)	63%	-	T2DM, BMI >25 kg/m^2
Zhou et al, 2016 (50)	Prospective, double-blinded, randomised, placebo-controlled	28	-	-	-	New T2DM
Nishimura et al. 2015 (45)	Prospective, double-blind, randomised, placebo-controlled	60	62.7 (±8.2)	78%	1.7%	Drug-naive T2DM, BMI ≤40 kg/m^2
Bailey et al, 2012 (51)	Prospective, double-blind, randomised, quadruple-arm, placebo-controlled	282	53.0 (±10.5)	50%	6.7% Unequal	T2DM, BMI ≤45.0 kg/m^2
Ferrannini et al, 2010 (52)	Prospective, randomised, single-arm pilot	274	52.2 (±10.7)	48%	4.0% Unequal	T2DM, BMI ≤ 45 kg/m^2

Legend

Study population characteristics. (-) represents omission of data. Equality of loss-to-follow up refers to equal or unequal drop out between intervention arms of the study. T2DM = type 2 diabetes mellitus, NAFLD = non-alcoholic fatty liver disease, HF = heart failure, BMI = body mass index, NASH = non-alcoholic steatohepatitis.

Table 2a: Changes in serum inflammatory biomarkers

Study	Comparison	Absolute change from baseline [±SD] (% change)	Control-subtracted change from baseline (% change)	p value
CRP/ hs-CRP (mg/L)				
Iannantuoni et al, 2019 (48)	24 week empagliflozin 10mg vs baseline	Decrease	-	<0.05
Bosch et al 2019 (46)	6 week 2mg empagliflozin vs placebo and baseline	-0.11 [±0.43] (-5.2%)	+0.11 (+5.3%)	0.583
Sezai et al 2019 (40)	12 month CRP canagliflozin 100mg vs baseline	-0.74 [±1.47] (-17.6%)	-	>0.05

Study	Comparison	Absolute change from baseline [±SD] (% change)	Control-subtracted change from baseline (% change)	p value
	12 month hs-CRP canagliflozin 100mg vs baseline	-0.18 [±0.46] (-46.2%)	-	<0.05
Noda et al 2019 (42)	14 day canagliflozin 100mg and teneligliptin vs baseline and standard care (teneligliptin)	-0.72 [±2.16] (-28.2%)	-0.29 (-11.3%)	>0.05
Eriksson et al, 2018 (36)	12 week dapagliflozin 10mg vs baseline and placebo	-0.23 (-9.3%)	-0.39 (-15.8%)	>0.05
Hattori, 2018 (33)	12 month empagliflozin 10mg vs baseline and placebo	-0.23 [±0.95] (-55.6%)	-0.99 (-74.4%)	<0.05
Garvey et al, 2018 (30)	52 week canagliflozin 300mg vs baseline and glimepiride 6mg or 8mg	-0.2 (-6.9%)	-0.3 (-10.3%)	-
Osonoi et al 2018 (43)	12 week 100mg Canagliflozin vs baseline	+0.2 [±0.89] (+22.2%)	-	0.515
Bouchi et al, 2017 (37)	12 month luseogliflozin 2.5mg vs baseline	-0.19 [±0.56] (-19.4%)	-	0.392
Tobita et al, 2017 (38)	24 week dapagliflozin 5mg vs baseline	-0.14* (-53.8%)	-	<0.01
Okamoto et al, 2016 (35)	12 week dapagliflozin 5mg vs baseline	-0.8 [±2.38] (-33.3%)	-	<0.01
Ferrannini et al, 2010 (52)	24 week 2.5, 5 or 10mg dapagliflozin, morning or night vs baseline and placebo	-	-1.53 [±17.5] to -2.67 [±18.2]	>0.05
Adiponectin (µg/ml)				
Aso et al 2019 (39)	24 week dapagliflozin 5mg/day vs baseline and standard care	+0.54 (+50.0%)	+0.60 (+56.0%)	<0.001
Eriksson et al, 2018	12 week dapagliflozin 10mg vs baseline and placebo	-298	-166	>0.05

Study	Comparison	Absolute change from baseline [±SD] (% change)	Control-subtracted change from baseline (% change)	p value
(36)		(-6.0%)	(-3.3%)	
Garvey et al, 2018 (30)	52 week canagliflozin 300mg vs baseline and glimepiride 6mg or 8mg	+1.1 (+31.4%)	+0.6 (+17.1%)	-
Bouchi et al, 2017 (37)	12 month luseogliflozin 2.5mg vs baseline	-0.3 [±4.74] (-3.4%)	-	0.233
Tobita et al, 2017 (38)	24 week dapagliflozin 5mg vs baseline	+1.6* (+22.9%)	-	<0.01
Matsumura et al, 2017 (34)	7 day canagliflozin 100mg vs baseline	+0.52* (+9.4%)	-	<0.05
Okamoto et al, 2016 (35)	12 week dapagliflozin 5mg vs baseline	+1.5 [±2.07] (+33.3%)	-	<0.01
Bailey et al, 2012 (51)	24 week dapagliflozin 1mg vs baseline and placebo	+0.72 [±1.08] (+11.3%)	+0.28 (+4.4%)	-
	24 week dapagliflozin 2.5mg vs baseline and placebo	+0.74 [±1.31] (+11.5%)	+0.30 (4.7%)	-
	24 week dapagliflozin 5mg vs baseline and placebo	+0.98 [±1.55] (+16.1%)	+0.49 (+8.1%)	-
Serum TNF-α (pg/ml)				
Garvey et al, 2018 (30)	52 week canagliflozin 300mg vs baseline and glimepiride 6mg or 8mg	+0.1 (+4.5%)	+0.2 (+9.0%)	-
Tan and Tan, 2018 (47)	6 month 10mg empagliflozin vs baseline	-15.0 [±13.7] (-37.4%)	-	0.002
	Then another 6 months 100mg canagliflozin vs baseline	-13.9 [±16.9] (-34.7%)	-	0.009

Study	Comparison	Absolute change from baseline [±SD] (% change)	Control-subtracted change from baseline (% change)	p value
Sato et al, 2018 (41)	6 month dapagliflozin (unspecified dose) vs baseline and standard treatment	-0.5 (-20.8%)	-0.53 (-22.1%)	<0.05
Matsumura et al, 2017 (34)	7 day canagliflozin 100mg vs baseline	-0.52* (-22.5%)	-	0.10
IL6 (pg/ml)				
Heerspink et al, 2019 (31)	104 week Canagliflozin 100mg vs Glimepiride	-	(-26.3%)[$] [-41.7, -6.7]	0.011
	104 week Canagliflozin 300mg vs Glimepiride	-	(-26.6%)[$] [-42.0, -7.2]	0.010
Dekkers et al 2018 (32)	12 week dapagliflozin 10mg vs placebo and baseline	(-24.0%)* [-37.9, -7.0] [†]	(-23.3%)	0.01
Garvey et al, 2018 (30)	52 week canagliflozin 300mg vs baseline and glimepiride 6mg or 8mg	-0.3 (-15.0%)	-0.5 (-25.0%)	-
Tan and Tan, 2018 (47)	6 months 10mg empagliflozin vs baseline	-10.5 [±8.97] (-52.0%)	-	0.022
	Then another 6 months 100mg canagliflozin vs baseline	-9.6 [±9.30] (-47.5%)	-	0.011
Bouchi et al, 2017 (37)	12 month luseogliflozin 2.5mg vs baseline	-0.66 [±0.68] (-33.0%)	-	0.278

Table 2b: Changes in oxidative stress biomarkers

Study	Comparison	Absolute change from baseline [±SD] (% change)	Control-subtracted change from baseline (% change)	p value

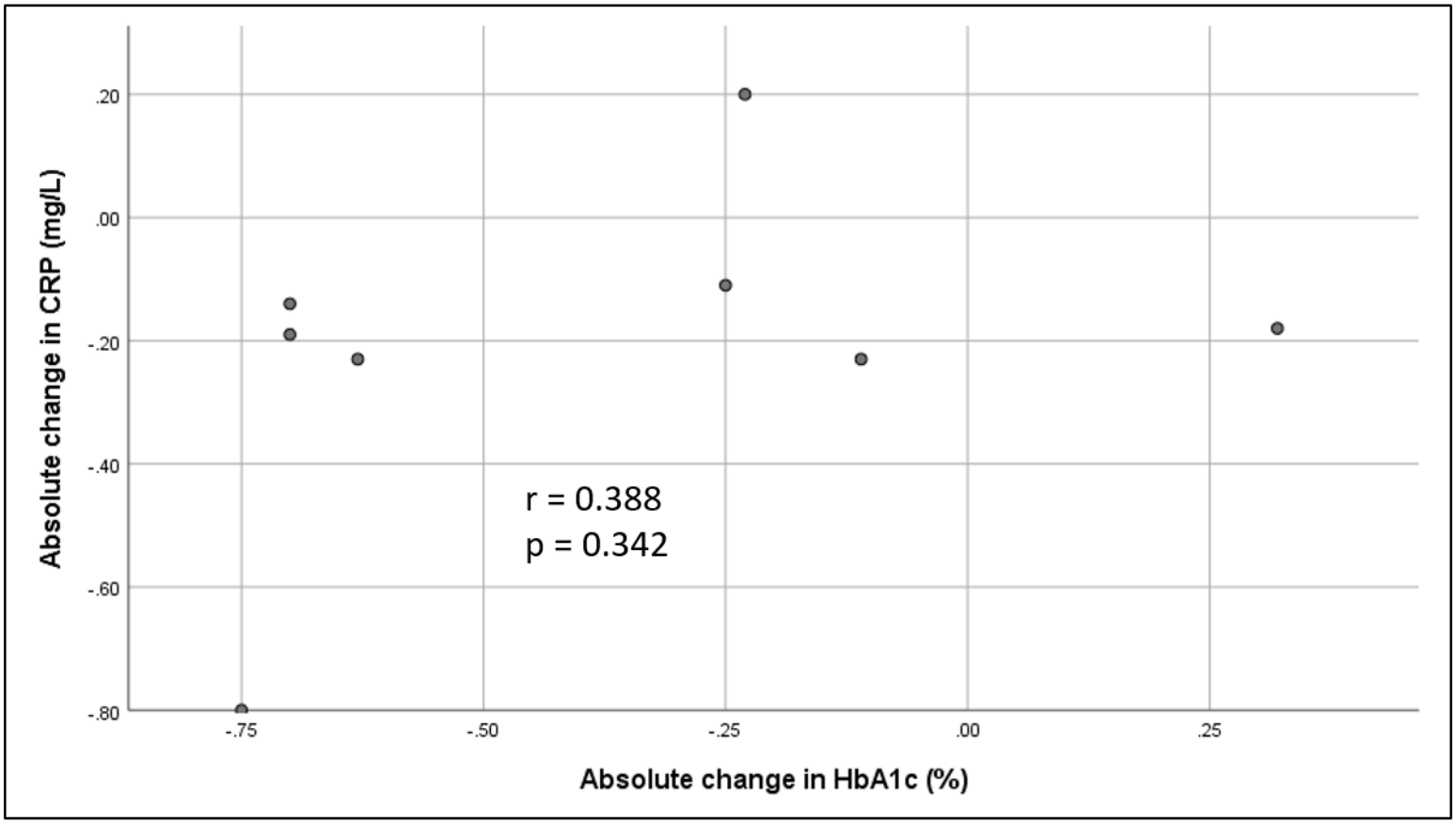
Study	Comparison	Absolute change from baseline [±SD] (% change)	Control-subtracted change from baseline (% change)	p value
8-iso-PGF2α				
Eriksson et al, 2018 (36)	12 week dapagliflozin 10mg/day vs placebo Urinary (ng/mg creatinine)	+0.001 [±0.022] (+1.4%)	+0.009 (+12.3%)	>0.05
Solini et al, 2017 (49)	2 days after single 10mg dapagliflozin treatment 24-hr Urinary (pg/ml)	-502 [±1224] (-30.3%)	-	0.04
Zhou et al, 2016 (50)	Dapagliflozin (between 5-10mg) vs placebo Serum (pg/mL)	Decrease	-	0.034
Nishimura et al, 2015 (45)	Empagliflozin 10mg vs placebo - 28 day urinary 8-iso-PGF2α in fasting state (pg/mL)	-48.1 [±181] (+24.7%)	-88.6 (+45.5%)	-
	Empagliflozin 25mg vs placebo- 28 day urinary 8-iso-PGF2α in fasting stage (pg/mL)	-33.5 [±184] (-22.9%)	-74.0 (-50.5%)	-
	Empagliflozin 10mg vs placebo, 28 day urinary 8-iso-PGF2α 24 hours after drug administration (pg/mL)	-28.4 [±80.3] (-20.5%)	-24.7 (-17.9%)	-
	Empagliflozin 25mg vs placebo, 28 day urinary 8-iso-PGF2α 24 hours after drug administration (pg/mL)	-46.8 [±81.7] (-31.5%)	-43.1 (-29.0%)	-
8-OHdG				
Shigiyama et al, 2017 (44)	16 week dapagliflozin 5mg/day + Metformin 750mg/day vs Metformin 1500mg/day Urinary (ng/mg Creatinine)	-0.6 [±1.80] (-13.0%)	-1.6 (-34.8%)	<0.001
Matsumura et al, 2017	Day 7 canagliflozin 100 mg/day vs baseline	-0.6	-	<0.05

Study	Comparison	Absolute change from baseline [±SD] (% change)	Control-subtracted change from baseline (% change)	p value
(34)	Urinary (ng/mg Creatinine)	(-5.3%)		

Legend

Serum biomarkers of inflammation (Table 1a) and direct biomarkers of oxidative stress (Table 1b). (-) represents information being unavailable or not relevant due to study design. SD represents the standard deviation of absolute mean average change in a biomarker from baseline, this was estimated where not directly available. % change represents the percentage change of a biomarker from baseline. * = median, † = 25th to 75th percentile, $ = least mean square percentage change. CRP = c-reactive protein, TNF α = tumour factor alpha necrosis, IL6 = interleukin-6, hsCRP = high-sensitivity c-reactive protein, 2,3-dinor-8-iso-PGF2α = 2,3-dinor-8-iso-prostaglandin F2α, 8-OHdG = 8-hydroxy-2' –deoxyguanosine.

Figure 2



Legend

Absolute change in CRP compared with absolute change in HbA1c from baseline to follow-up. The Pearson's correlation coefficient (r) is displayed.

Supplementary tables

Supplementary table 1a – comprehensive overview of changes in inflammatory biomarkers

Study	Type of Study	n	Comparison and Study Population	Outcome (serum)	Baseline control	Baseline SGLT2i	Follow-up Control	Follow-up SGLT2i	Percentage Change					
									Baseline vs control	p value	Baseline vs SGLT2i	p value	Control vs SGLT2i	p value
Iannantuoni et al, 2019 (48)	Observational, prospective, open-label study	17	Empagliflozin 10 mg vs baseline Long-standing (>10 years) T2DM patients aged between 40-70.	24 week hsCRP (mg/L)	-	-	-	Decreased	-	< 0.05	-	-	-	-
				24 week IL10 (pg/mL)	-	-	-	Decreased	-	< 0.05	-	-	-	-
Bosch et al 2019 (46)	Prospective, double-blind, randomized, placebo-controlled trial	58	25 mg Empagliflozin vs placebo and baseline T2DM patients with an eGFR ≥ 60 ml/min/1.73 m^2 (mean SD) *excludes patients with CRP >5 mg/L	6 week hsCRP (mg/L)	2.10 (1.72)		1.88 (1.32)	1.99 (1.19)	- 10.5%	0.283	- 5.2%	0. 583	+ 5.3%	0.458
Heerspink et al, 2019 (31)	Retrospective analysis from a prospective randomised double-blind, controlled trial (CANTATA-SU trial)	296	Canagliflozin 100 mg and 300 mg vs Glimepiride as least mean square T2DM patients with urinary albumin/creatinine ratio >1.7	104 week TNFR1 100 mg	-	-	-	-	-	-	-	-	- 5.9%	0.013
				104 week TNFR1 300 mg	-	-	-	-	-	-	-	-	- 9.2%	< 0.001
				104 week IL6 100 mg	-	-	-	-	-	-	-	-	- 26.3%	0.011
				104 week IL6 300 mg	-	-	-	-	-	-	-	-	- 26.6%	0.010
				104 week MMP7 100 mg	-	-	-	-	-	-	-	-	- 16.3%	0.110
				104 week MMP7 300 mg	-	-	-	-	-	-	-	-	- 24.9%	0.011
				104 week MMP8 100 mg	-	-	-	-	-	-	-	-	- 7.4%	0.360

Study	Type of Study	n	Comparison and Study Population	Outcome (serum)	Baseline control	Baseline SGLT2i	Follow-up Control	Follow-up SGLT2i	Percentage Change					
									Baseline vs control	p value	Baseline vs SGLT2i	p value	Control vs SGLT2i	p value
				104 week MMP8 300 mg	-	-	-	-	-	-	-	-	- 15.5%	0.070
				104 week fibronectin 1 100 mg	-	-	-	-	-	-	-	-	- 15.8	0.040
				104 week fibronectin 1 300 mg	-	-	-	-	-	-	-	-	- 14.9	0.055
Aso et al 2019 (39)	Prospective, randomised, open-label, blinded endpoint trial	57	Dapagliflozin 5 mg/day vs baseline and standard care T2DM patients with NAFLD (median + IQR)	24 week ferritin (n/mL)	79.5 (32.4, 150)	63.0 (30.0, 157.8)	46.5 (22.6, 106)	76.7 (36.3, 116.0)	+ 21.7%	0.727	- 41.5%	<0.001	- 63.2%	-
				24 week TGF-β1 (ng/mL)	1.29 (0.97, 1.81)	1.32 (1.00, 2.29)	1.20 (0.98, 1.45)	1.11 (0.98, 1.51)	- 7.0%	0.229	- 15.9%	0.016	- 8.9%	-
				24 week HMW adiponectin (µg/mL)	1.31 (0.44, 2.53)	1.08 (056, 4.11)	1.79 (0.39, 2.55)	1.62 (0.91, 4.67)	+ 36.6%	0.389	+ 50.0%	< 0.001	+ 13.4	-
Sezai et al 2019 (40)	Prospective, randomised, open-label, add-on, controlled trial (CANOSSA trial)	35	Canagliflozin 100 mg vs baseline Chronic T2DM patients with heart failure (mean SD) *SD calculated from SEM	12 month CRP (mg/L)	-	4.21 (2.07)	-	3.47 (1.72)	-	-	- 17.6%	> 0.05	-	-
				12 month Hs-CRP (mg/L)	-	0.39 (0.41)	-	0.21 (0.24)	-	-	- 46.2%	< 0.05	-	-
				12 month RLP-cho (mg/dL)	-	5.89 (5.56)	-	3.69 (3.67)	-	-	- 37.4%	< 0.05	-	-
				12 month EPA/AA ratio	-	0.52 (0.41)	-	0.57 (0.35)	-	-	+ 9.6%	> 0.05	-	-
Noda et al 2019 (42)	Observational, open-label study	12	Canagliflozin 100 mg and teneligliptin vs baseline and standard care (teneligliptin) T2DM patients (mean SD)	14 day hs-CRP (mg/L)	2.57 (2.72)		2.14 (2.93)	1.85 (2.38)	- 16.7%	-	- 28.2%	-	- 13.8%	> 0.05

Study	Type of Study	n	Comparison and Study Population	Outcome (serum)	Baseline control	Baseline SGLT2i	Follow-up Control	Follow-up SGLT2i	Percentage Change					
									Baseline vs control	p value	Baseline vs SGLT2i	p value	Control vs SGLT2i	p value
Dekkers et al 2018 (32)	Prospective, randomised, double-blind, placebo-controlled cross-over clinical trial	31	Dapagliflozin 10 mg vs placebo and baseline T2DM patients with alb/cr ratio > 100 mg/g + < 3500 mg/g (median pg/24h + 25th to 75th percentile)	12 weeks total IgG (pg/24 h)	2269 (875-4600)		-	-	+ 4.3% (-12.4, 24.2)	0.640	- 25.3% (-38.1, - 9.9)	0.010	- 29.6	-
				12 weeks total IgG4 (pg/24 h)	4 (1-8)		-	-	+ 3.6% (-21.0, 36.0)	0.800	- 32.2% (-49.1, - 9.7)	0.010	- 35.8	-
				12 weeks IL6 (pg/24 h)	3 (2-5)		-	-	- 0.7% (-18.1, 20.5)	0.950	- 24.0% (-37.9, - 7.0)	0.010	- 23.3%	-
Eriksson et al, 2018 (36)	Multicentre, prospective, randomised placebo-controlled double-blind double-dummy four-armed parallel-group trial (EFFECT-II trial)	42	Dapagliflozin 10 mg vs baseline and placebo Overweight T2DM patients with NAFLD (mean SD)	12 week CRP (mg/dL)	1.88 (1.89)	2.47 (1.96)	2.04	2.24	+ 8.5%	> 0.05	- 9.3%	> 0.05	- 17.8%	-
				12 week adiponectin (µg/L)	5591 (3798)	4978 (3142)	5459	4680	- 2.4%	> 0.05	- 6.0%	> 0.05	- 3.6%	-
				12 week leptin (µg/L)	16.8 (15.9)	15.5 (13.3)	17.2	15.1	+ 2.3%	> 0.05	- 2.9%	> 0.05	- 5.2%	-
				12 week osteopontin (ng/ml)	60.6 (23.3)	69.2 (±46.9)	51.6	56.8	- 14.8%	> 0.05	- 17.9%	> 0.05	- 3.1%	-
Hattori, 2018 (33)	Single centre, open-label, randomised, prospective study	102	Empagliflozin 10 mg vs baseline and placebo T2DM patients at 3, 6, 9 and 12 months (mean SD)	12 month CRP (mg/L)	1.46 (1.4)	1.33 (1.0)	1.71 (1.64)	0.59 (0.42)	+ 17.1%	> 0.05	- 55.6%	< 0.05	- 72.7%	0.007
				12 month RLP-C (mg/dL)	6.27 (3.96)	8.13 (5.02)	7.91 (5.57)	3.94 (2.10)	+ 26.2%	> 0.05	- 51.5%	< 0.05	- 77.7%	0.029
Garvey et al, 2018 (30)	Phase 3, randomised, prospective, double-blind, active-controlled trial	100	Canagliflozin 300 mg vs baseline and glimepiride 6mg or 8mg T2DM patients (median +IQR) *Excludes patients with CRP >10 mg/L	52 week CRP (µg/mL)	2.9 (1.4, 5.0)	2.9 (1.4, 5.7)	3.0	2.7	+ 3.4%	-	- 6.9%	-	- 10.3%	-
				52 week TNFα (pg/ml)	2.2 (1.8, 2.6)	2.2 (1.8, 2.5)	2.1	2.3	- 4.5%	-	+ 4.5%	-	+ 9.1%	-
				52 week IL6 (pg/ml)	1.8 (1.1, 2.9)	2.0 (1.3, 2.9)	2.0	1.7	+11.1%	-	- 15.0%	-	- 26.1%	-
				52 week VCAM-1 (ng/ml) (LS mean change SD)	710.6 (210.1)	711.9 (165.9)	706.7	729.7	+0.5%	-	+ 2.5%	-	+ 1.6%	-

Study	Type of Study	n	Comparison and Study Population	Outcome (serum)	Baseline control	Baseline SGLT2i	Follow-up Control	Follow-up SGLT2i	Percentage Change					
									Baseline vs control	p value	Baseline vs SGLT2i	p value	Control vs SGLT2i	p value
				52 week adiponectin (µg/ml)	3.0 (2.2, 4.3)	3.5 (2.5,4.7)	3.0	4.1	0%	-	+ 17.1%	-	17.1%	-
				52 week leptin (ng/ml)	11.0 (5.8, 19.0)	13.0 (7.8, 20.0)	12.0	11.9	+ 9.1%	-	- 8.5%	-	- 17.6%	-
Osonoi et al 2018 (43)	Observational, single-arm, open label study	20	100 mg Canagliflozin vs baseline T2DM patients with microalbuminuria (mean SD)	12 week CRP (mg/L)	-	0.9 (1.3)	-	1.1 (±1.6)	-	-	+ 22.2%	0.515	-	-
				12 week TNFR1 (pg/mL)	-	1256 (217)	-	1220 (186)	-	-	- 2.8%	0.282	-	-
				12 week TNFR2 (pg/mL)	-	2587 (600)	-	2826 (605)	-	-	+ 9.2%	0.047	-	-
				12 week IL-18 (pg/mL)	-	313 (111)	-	321 (106)	-	-	+ 2.6%	0.705	-	-
Tan and Tan, 2018 (47)	Conference abstract	32	6 months 100 mg canagliflozin then 6 months 10 mg empagliflozin + 6 months 10 mg empagliflozin then 6 months 100 mg canagliflozin vs baseline Male T2DM patients (unspecified dispersion statistics)	6 month empa TNF-α (pg/ml)	-	40.1 (6.7)	-	25.1 (3.8)	-	-	- 37.4%	0.002	-	-
				12 month cana then empa TNF-α (pg/ml)	-	40.1 (6.7)	-	26.2 (4.9)	-	-	- 34.7%	0.009	-	-
				6 month empa IL6 (pg/ml)	-	20.2 (8.3)	-	9.7 (3.4)	-	-	- 52.0%	0.022	-	-
				12 month cana then empa IL6 (pg/ml)	-	20.2 (8.3)	-	10.6 (4.2)	-	-	- 47.5%	0.011	-	-

Study	Type of Study	n	Comparison and Study Population	Outcome (serum)	Baseline control	Baseline SGLT2i	Follow-up Control	Follow-up SGLT2i	Percentage Change					
									Baseline vs control	p value	Baseline vs SGLT2i	p value	Control vs SGLT2i	p value
				6 month empa IFN-γ (pg/ml)	-	23.3 (4.5)	-	10.1 (2.4)	-	-	- 56.7%	0.001	-	-
				12 month cana then empa IFN-γ (pg/ml)	-	23.3 (4.5)	-	11.0 (1.4)	-	-	- 52.8%	0.007	-	-
Sato et al, 2018 (41)	Single-centre, randomised, prospective, controlled trial (no mention of label blinding)	40	Dapagliflozin (unspecified dose) vs baseline and standard treatment T2DM patients with coronary artery disease (mean SD)	6 month TNF-α (pg/ml)	2.20 (0.7)	2.40 (0.7)	2.23	1.90	1.4%	>0.1	- 20.8%	<0.05	- 22.2%	0.03
Bouchi et al, 2017 (37)	Randomised, prospective, single-arm pilot controlled trial	19	Luseogliflozin 2.5 mg vs baseline T2DM patients with HbA1c 6.5-9 + BMI >25 (mean unspecified dispersion statistics) *converted from log to original data	12 month CRP (mg/L)	-	0.98 (0.35)	-	0.79 (0.47)	-	-	- 19.4%	0.392	-	-
				12 month IL6 (pg/ml)	-	2.00 (1.70)	-	1.34 (1.82)	-	-	- 33%	0.278	-	-
				12 month Leptin (ng/ml)	-	13.9 (6.1)	-	13.1 (6.8)	-	-	- 5.8%	0.377	-	-
				12 month Adiponectin (μg/ml)	-	8.8 (3.3)	-	8.5 (3.4)	-	-	- 3.4%	0.233	-	-

Study	Type of Study	n	Comparison and Study Population	Outcome (serum)	Baseline control	Baseline SGLT2i	Follow-up Control	Follow-up SGLT2i	Percentage Change					
									Baseline vs control	p value	Baseline vs SGLT2i	p value	Control vs SGLT2i	p value
Tobita et al, 2017 (38)	Prospective, observational, open-label, uncontrolled pilot study	11	Dapagliflozin 5 mg vs baseline T2DM patients with NASH (median + IQR)	24 week hs-CRP (mg/L)	-	0.26 (0.11-0.53)	-	0.12 (0.05-0.32)	-	-	- 54%	<0.01	-	-
				24 week Adiponectin (μg/ml)	-	5.40 (4.60-8.85)	-	7.00 (5.60-11.80)	-	-	+ 22.9%	<0.01	-	-
Matsumura et al, 2017 (34)	Prospective, observational, open-label study	15	Canagliflozin 100 mg vs baseline T2DM patients with metabolic syndrome, hypertension and/or dyslipidaemia (median + graphical IQR)	7 day TNF-α (pg/ml)	-	2.31	-	1.79	-	-	- 22.5%	0.10	-	-
				7 day Adiponectin (μg/ml)	-	5.01	-	5.53	-	-	+ 9.4%	<0.05	-	-
Okamoto et al, 2016 (35)	Prospective, observational, single-arm, open-label study	27	Dapagliflozin 5 mg vs baseline Overweight T2DM patients (mean SD)	12 week hs-CRP (mg/L)	-	2.41 (2.81)	-	1.61 (1.96)	-	-	- 33.3%	<0.01	-	-
				12 week Adiponectin (μg/ml)	-	5.1 (2.3)	-	6.7 (4.2)	-	-	+ 33.3%	<0.01	-	-
Bailey et al, 2012 (51)	Phase 3, randomised, double-blind, quadruple-arm, placebo-controlled study	282	Dapagliflozin 1, 2.5 and 5 mg vs baseline and placebo Antidiabetic naïve T2DM patients (mean SD)	24 week Leptin (μg/L) 1 mg	26.8 (24.5)	25.06 (23.6)	25.3 (24.3)	23.0 (18.9)	- 4.5%	-	- 8.7%	-	- 4.2%	-
				24 week Leptin (μg/L) 2.5 mg	26.8 (24.5)	22.24 (19.15)	25.3 (24.3)	21.0 (19.8)	- 4.5%	-	- 8.9%	-	- 4.4%	-
				24 week Leptin (μg/L) 5 mg	26.8 (24.5)	27.9 (31.2)	25.3 (24.3)	22.9 (24.7)	- 4.5%	-	- 15.9%	-	- 11.4%	-
				24 week Adiponectin (μg/ml) 1 mg	6.61 (3.38)	6.39 (2.95)	7.05 (3.35)	7.11 (3.19)	+ 7.2%	-	+ 11.3%	-	+ 4.1	-
				24 week Adiponectin (μg/ml) 2.5 mg	6.61 (3.38)	6.44 (2.95)	7.05 (3.35)	7.18 (3.51)	+ 7.2%	-	+ 11.6%	-	+ 4.4%	-
				24 week Adiponectin (μg/ml) 5 mg	6.61 (3.38)	6.08 (2.71)	7.05 (3.35)	7.01 (3.25)	+ 7.2%	-	+ 14.6%	-	+ 7.4%	-
Ferrannini et al, 2010 (52)	Phase 3, randomised, prospective, parallel-group, double-blind, placebo-controlled trial	274	2.5, 5 or 10 mg dapagliflozin, morning or night vs baseline and placebo Antidiabetic naïve T2DM patients (placebo-subtracted adjusted mean ±SEM) *assumed only main cohort as unspecified	24 week hs-CRP (mg/L)	-	-	-	-1.53 [±17.5] to -2.67 [±18.2]	-	-	-	-	- 74.5%	-

Supplementary table 1b – comprehensive overview of changes in oxidative stress biomarkers

Study	Type of Study	n	Comparison and Study Population	Outcome Measure	Baseline Control	Baseline SGLT2i	Follow-up Control	Follow-up SGLT2i	Percentage Change					
									Baseline vs Control	p value	Baseline vs SGLT2i	p value	Control vs SGLT2i	p value
Sezai et al, 2019 (40)	Prospective, randomised trial (CANOSSA trial)	35	Canagliflozin 100 mg/day Chronic T2DM patients with heart failure	3 month serum Ox-LDL (U/L)	-	-	-	-	-	-	Decrease	< 0.05	-	-
				6 month serum Ox-LDL (U/L)	-	-	-	-	-	-	Decrease	< 0.05	-	-
				12 month serum Ox-LDL (U/L)	-	-	-	-	-	-	Decrease	< 0.05	-	-
Iannantuoni et al, 2019 (48)	Observational, prospective, open-label study	17	Empagliflozin 10 mg/day Long-standing (>10 years) T2DM patients aged between 40-70.	12 week leukocyte mitochondrial superoxide production	-	-	-	-	-	-	Decrease	≥ 0.05	-	-
				24 week leukocyte mitochondrial superoxide production	-	-	-	-	-	-	Decrease	< 0.05	-	-
				12 week leukocyte mitochondrial glutathione content	-	-	-	-	-	-	Increase	< 0.05	-	-
				24 week leukocyte mitochondrial glutathione content	-	-	-	-	-	-	Increase	< 0.05	-	-
				12 week leukocyte mitochondrial glutathione s-reductase mRNA level	-	-	-	-	-	-	-	-	-	-
				24 week leukocyte mitochondrial glutathione s-reductase mRNA level	-	-	-	-	-	-	Increase	< 0.05	-	-
				12 week leukocyte mitochondrial catalase mRNA level	-	-	-	-	-	-	-	-	-	-
				24 week leukocyte mitochondrial catalase mRNA level	-	-	-	-	-	-	Increase	< 0.05	-	-
Eriksson et al, 2018 (36)	Randomised, placebo-controlled, double-blind study	42	Dapagliflozin 10 mg/day vs placebo	12 week serum acetylcarnitine (μmol/l)	10.36 (±3.64)	9.14 (±2.50)	-	-	- 3.3% (±20.5%)	≥ 0.05	+ 12.7% (±23.5%)	≥ 0.05	+ 16.0%	≥ 0.05

Study	Type of Study	n	Comparison and Study Population	Outcome Measure	Baseline Control	Baseline SGLT2i	Follow-up Control	Follow-up SGLT2i	Percentage Change					
									Baseline vs Control	p value	Baseline vs SGLT2i	p value	Control vs SGLT2i	p value
	(EFFECT-II study)		Stable patients with T2DM and NAFLD aged 40-75 years. (mean ±SD)	12 week serum 2-hydroxynonenal (pg/ml)	5.74 (±1.14)	5.51 (±1.57)	-	-	- 3.0% (±12.0%)	≥ 0.05	- 1.5% (±15.2%)	≥ 0.05	+ 1.5%	≥ 0.05
				12 week serum 2-hydroxyhexanal (pg/ml)	3.76 (±1.35)	3.30 (±0.69)	-	-	- 0.8% (±30.6%)	≥ 0.05	+ 3.3% (±17.6%)	≥ 0.05	+ 4.1%	≥ 0.05
				12 week urinary 8-iso-PGF2α (ng/mg creatinine)	0.088 (±0.068)	0.073 (±0.024)	-	-	- 9.1% (±37.5%)	≥ 0.05	+ 1.4% (±30.1%)	≥ 0.05	+10.6%	≥ 0.05
				12 week urinary 2,3-dinor-8-iso-PGF2α (ng/mg creatinine)	1.58 (±0.79)	1.43 (±0.70)	-	-	+ 7.0% (±27.2%)	≥ 0.05	+ 25.9% (±45.5%)	≥ 0.05	+ 18.9	≥ 0.05
Solini et al, 2017 (49)	Prospective observational trial	16	Dapagliflozin 10 mg Outpatients with T2DM aged 40-70 years. (mean ±SD)	2 days after single dapagliflozin treatment 24-hr urinary 8-iso-PGF2α (pg/ml)	-	1659 (±1029)	-	1157 (±663)	-	-	- 30.3%	0.04	-	-
Shigiyama et al, 2017 (44)	Prospective, randomised, open-label, blinded-endpoint, parallel-group, comparative clinical trial (DEFENCE study)	80	Dapagliflozin 5mg/day + Metformin 750 mg/day vs5. Metformin 1500 mg/day T2DM patients treated with 750 mg/day Metformin (mean ±SD)	16 week urinary 8-OHdG/creatinine (ng/mg Cre)	4.8 (±2.0)	4.6 (±2.4)	5.8 (±2.3)	4.0 (±1.9)	+ 22.9% (±45.8%)	-	- 13.0% (±39.1%)	>0.05	- 36.2%	< 0.001
Matsumura et al, 2017 (34)	Prospective observational trial	15	Canagliflozin 100 mg/day from Day 4 onwards. T2DM patients who had received insulin therapy for 1 year or more	Day 7 urinary 8-OHdG (ng/mg Cre)	-	11.4	-	10.8	-	-	-5.3%	< 0.05	-	-

Study	Type of Study	n	Comparison and Study Population	Outcome Measure	Baseline Control	Baseline SGLT2i	Follow-up Control	Follow-up SGLT2i	Percentage Change					
									Baseline vs Control	p value	Baseline vs SGLT2i	p value	Control vs SGLT2i	p value
Zhou et al, 2016 (50)	Randomised, double-blinded, placebo-controlled, parallel grouped study	28	Dapagliflozin vs Placebo (dose unclear, between 5-10mg) Newly diagnosed T2DM	24 week serum 8-iso-PGF2α (pg/mL)	-	-	-	-	-	-	Decrease	0.034	-	-
Nishimura et al, 2015 (45)	Randomised, double-blind, placebo-controlled, parallel-group study	60	Empaglifllozin 10 mg vs Empagliflozin 25 mg vs Placebo Stable T2DM patients aged 40-74 years with a BMI <40. (adjusted mean ±SE or 95% CI)	Empa 10 mg - 28 day urinary 8-iso-PGF2α in fasting state (pg/mL)	197.8 (±27.0)	194.6 (±29.4)	-	-	+ 20.5% (±11.4%)	-	- 24.7% (±12.0%)	-	- 45.2% (- 78.2%, - 11.5%)	0.010
				Empa 25 mg - 28 day urinary 8-iso-PGF2α in fasting stage (pg/mL)	197.8 (±27.0)	146.5 (±18.5)	-	-	+ 20.5% (±11.4%)	-	- 22.9% (±16.2%)	-	- 43.0% (- 70.7%, - 4.1%)	0.028
				Empa 10 mg - 28 day urinary 8-iso-PGF2α 24 hours after drug administration (pg/mL)	115.5 (±11.0)	138.3 (±20.6)	-	-	- 3.2% (±8.9%)	-	- 20.5% (±7.5%)	-	- 19.5% (- 47.2%, + 4.5%)	0.103
				Empa 25 mg - 28 day urinary 8-iso-PGF2α 24 hours after drug administration (pg/mL)	115.5 (±11.0)	148.6 (±21.7)	-	-	- 3.2% (±8.9%)	-	- 31.5% (±7.1%)	-	- 32.6% (- 63.1%, - 8.9%)	0.006

Legend

Serum biomarkers of inflammation (Table 1a) and direct biomarkers of oxidative stress (Table 1b). (-) represents information being unavailable or not relevant due to study design. CRP = c-reactive protein, TNF α = tumour necrosis, IL6 = interleukin-6, IL10 = interleukin-10, IL18 = interleukin-18, factor alpha, hsCRP = high-sensitivity c-reactive protein,TNFR1 and 2 = tumor necrosis factor receptor 1 and 2, MMP7 and 8 = matrix metalloprotease 7 and 8, TGF-β1 = transforming growth factor beta 1, HMW adiponectin =

high-molecular weight adiponectin, RLP-cho = remnant lipoprotein cholesterol, EPA/AA = eicosapentaenoic acid to arachidonic acid ratio, IgG = immunoglobulin G, VCAM-1 = vascular adhesion molecule 1, IFN-γ = interferon gamma, Ox-LDL = oxidised low-density lipoprotein, mRNA = messenger ribonucleic acid, 8-iso-PGF2α = 8-iso-prostaglandin F2α, 2,3-dinor-8-iso-PGF2α = 2,3-dinor-8-iso-prostaglandin F2α, 8-OHdG = 8-hydroxy-2' –deoxyguanosine, SD = standard deviation, 95% CI = 95% confidence interval, SEM = standard error of the mean, IQR = interquartile range, T2DM = type 2 diabetes mellitus, CRP = c-reactive protein, NAFLD = non-alcoholic fatty liver disease and BMI = body mass index.

Supplementary table 2 - Grading of Recommendations Assessment, Development and Evaluation (GRADE) system

Certainty Assessment							№ of patients	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations			
Follow-up serum C-Reactive Protein (CRP)									
12	7 randomised trials	serious [a]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	630	⨁⨁⨁◯ MODERATE	CRITICAL
	5 observational studies	serious [c]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	102	⨁⨁◯◯ LOW	CRITICAL
Follow-up serum Adiponectin									

Certainty Assessment							№ of patients	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations			
8	5 randomised trials	serious [d]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	500	⨁⨁⨁◯ MODERATE	CRITICAL
	3 observational studies	serious [e]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	53	⨁⨁◯◯ LOW	CRITICAL
Follow-up serum Interleukin-6 (IL6)									

Certainty Assessment							№ of patients	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations			
4	3 randomised trials	serious [f]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	315	⨁⨁⨁◯ MODERATE	CRITICAL
	1 observational study	serious [g]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	32	⨁⨁◯◯ LOW	CRITICAL
Tumour Necrosis Factor alpha (TNF-α)									

Certainty Assessment							№ of patients	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations			
4	2 randomised trials	serious [h]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	140	⨁⨁⨁◯ MODERATE	CRITICAL
	2 observational studies	serious [i]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	47	⨁⨁◯◯ LOW	CRITICAL
8-iso-prostaglandin F2α (8-iso-PGF2α)									

Certainty Assessment							№ of patients	Certainty	Importance
№ of studies	Study design	Risk of bias	Inconsistency	Indirectness	Imprecision	Other considerations			
4	3 randomised trials	not serious [j]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	130	⊕⊕⊕⊕ HIGH	CRITICAL
	1 observational study	not serious [k]	not serious	not serious	serious [b]	all plausible residual confounding would suggest spurious effect, while no effect was observed	16	⊕⊕⊕⊕ HIGH	CRITICAL

Explanations

a) 1) Selection bias as Bosch et al 2019 recruited from local newspapers 2) performance and detection bias due to non-blinding in Aso et al 2019, Sezai et al 2019, Hattori 2018 and Bouchi et al 2017 3) attrition bias due to loss to follow up bias due to Heerspink et al 2019, Eriksson et al 2018, and potential loss to follow up due to Bosch et al 2019, Hattori 2018 and Okamoto et al 2016.

b) Wide standard deviations across results

c) 1) Selection bias due to non-randomisation of Noda et al 2019, Osonoi et al 2018, Tobita et al 2017 and Okamoto et al 2016 2) performance and detection bias due to non-blinding in Noda et al 2019, Osonoi et al 2018, Tobita et al 2017 and Okamoto et al 2016 3) attrition bias due to loss to follow up bias due to Noda et al 2019, Tobita et al 2017, and potential loss to follow up due to Okamoto et al 2016.

d) 1) Performance and detection bias due to non-blinding in Aso et al 2019, Bouchi et al 2017 2) attrition bias due to loss to follow up bias due to Eriksson et al 2018 and Bailey et al 2012.

e) 1) Selection bias due to non-randomisation of Tobita et al 2017, Matsumura et al 2017 and Okamoto et al 2016 2) performance and detection bias due to non-blinding in Tobita et al 2017, Matsumura et al 2017 and Okamoto et al 2016 3) attrition bias due to loss to follow up bias due to Tobita et al 2017, and potentially Matsumura al 2017 and Okamoto et al 2016.

f) 1) Performance and detection bias due to non-blinding in Bouchi et al 2017 2) attrition bias due to loss to follow up bias due to Heerspink et al 2019

g) 1) Selection bias due to non-randomisation of Osonoi et al 2018 and potential selection bias due to no mention of randomisation in Tan and Tan 2018 2) performance and detection bias due to non-blinding in Osonoi et al 2018, Bouchi et al 2017 and potentially Tan and Tan 2018.

h) 1) Potential bias due to no mention of blinding in Sato et al 2018 2) attrition bias due to loss to follow up bias due to Garvey et al 2018 and potentially Tan and Tan 2018 and Matsumura et al 2017.

i) 1) Potential selection bias due to no mention of randomisation in Tan and Tan 2018 2) performance and detection bias due to non-blinding in Matsumura et al 2017 3) attrition bias due to loss to follow up bias potentially due to Tan and Tan 2018 and Matsumura et al 2017.

j) 1) Potential attrition bias due to no mention of loss to follow up in Zhou et al 2016.

k) 1) Potential selection bias due to no mention of randomisation in Solini et al 2017.

Supplementary table 3a - Cochrane Risk-of-Bias Tool for Randomised trials (RoB 2)

First Author, Year (Ref. #)	Bias due to randomisation process	Bias in assignment and adherence to intervention	Bias due to Loss to follow up	Bias due to measurement of outcomes	Bias in selection of reported result	Overall risk of bias judgment
Follow-up serum C-Reactive Protein (CRP)						
Bosch, 2019	Low	Low	Unavailable	Low	Low	Some concerns
Sezai, 2019	Low	High	Low	Some concerns	Low	High
Eriksson, 2018	Low	Low	Low	Low	Low	Low
Hattori, 2018	Low	High	Unavailable	Some concerns	Low	Some concerns
Garvey, 2018	Low	Low	Some concerns	Low	Low	Some concerns
Bouchi, 2017	Low	Some concerns	Low	Some concerns	Low	Some concerns
Ferrannini 2010	Low	Low	Low	Low	Low	Low
Follow-up serum Adiponectin						
Aso, 2019	Low	Low	Low	Low	Low	Low

First Author, Year (Ref. #)	Bias due to randomisation process	Bias in assignment and adherence to intervention	Bias due to Loss to follow up	Bias due to measurement of outcomes	Bias in selection of reported result	Overall risk of bias judgment
Eriksson, 2018	Low	Low	Low	Low	Low	Low
Garvey, 2018	Low	Low	Some concerns	Low	Low	Some concerns
Bouchi, 2017	Low	Some concerns	Low	Some concerns	Low	Some concerns
Bailey 2012	Low	Low	High	Low	Low	High
Follow-up serum Interleukin-6 (IL6)						
Heerspink, 2019	Low	Low	Some concerns	Low	Low	Some concerns
Dekkers, 2018	Low	Low	Low	Low	Low	Low
Bouchi, 2017	Low	Some concerns	Low	Some concerns	Low	Some concerns
Follow-up serum Tumour Necrosis Factor alpha (TNF-α)						
Garvey 2018	Low	Low	Some concerns	Low	Low	Some concerns
Sato 2018	Low	Some concerns	Low	Low	Low	Some concerns
Oxidative stress biomarkers						

First Author, Year (Ref. #)	Bias due to randomisation process	Bias in assignment and adherence to intervention	Bias due to Loss to follow up	Bias due to measurement of outcomes	Bias in selection of reported result	Overall risk of bias judgment
Sezai, 2019	Low	Some concerns	Low	Some concerns	Low	High
Eriksson, 2018	Low	Low	Low	Low	Low	Low
Shigiyama, 2017	Low	Low	Low	Low	Low	Low
Zhou, 2016	Low	Low	Some concerns	Low	Low	Some concerns
Nishimura, 2015	Low	Low	Low	Low	Low	Low

Supplementary table 3b - Cochrane Risk-of-Bias Tool for Non-Randomised trials (ROBINS-I)

First author, Year (Ref. #)	Bias due to confounding	Bias in selection of participants	Bias in classification of interventions	Bias due to departures from intended interventions	Bias due to missing data	Bias due to measurement of outcomes	Bias in selection of reported results	Overall risk of bias judgment
Follow-up serum C-Reactive Protein (CRP)								
Iannantuoni, 2019	Low	Low	Low	Low	Serious	Moderate	Low	Serious
Noda, 2019	Low	Low	Low	Low	Serious	Low	Low	Serious
Osonoi, 2018	Moderate	Low	Low	Low	Low	Moderate	Low	Moderate

Tobita, 2017	Low	Low	Low	Low	Moderate	Moderate	Low	Moderate
Okamoto, 2016	Low	Low	Low	Low	Unavailable	Moderate	Low	Moderate
Follow-up serum Adiponectin								
Tobita, 2017	Low	Low	Low	Low	Moderate	Moderate	Low	Moderate
Matsumura, 2017	Moderate	Low	Low	Low	Unavailable	Moderate	Low	Moderate
Okamoto, 2016	Low	Low	Low	Low	Unavailable	Moderate	Low	Moderate
Follow-up serum Interleukin-6 (IL6)								
Tan and Tan, 2018	Unavailable	Moderate	Low	Low	Low	Moderate	Low	Moderate
Follow-up serum Tumour Necrosis Factor alpha (TNF-α)								
Tan and Tan, 2018	Unavailable	Moderate	Low	Low	Low	Moderate	Low	Moderate
Matsumura, 2017	Moderate	Low	Low	Low	Unavailable	Moderate	Low	Moderate
Oxidative stress biomarkers								
Iannantuoni, 2019	Low	Low	Low	Low	Serious	Moderate	Low	Serious
Solini, 2017	Low	Low	Low	Low	Low	Moderate	Low	Moderate
Matsumura, 2017	Moderate	Low	Low	Low	Unavailable	Moderate	Low	Moderate

Supplementary table 4 – co-morbidity and pharmacological intensity scores correlated with change in CRP

Study	Participant co-morbidity from inclusion criteria	Co-morbidity score	Pharmacological intensity from inclusion criteria	Pharmacological intensity score
Bosch et al	Nil	0	4 months antidiabetic medication free.	0
Sezai et al	Nil	0	Any concomitant oral antidiabetic medication	2
Noda et al	Nil	0	metformin only or no antidiabetic medications.	1
Eriksson et al	NAFLD + overweight	9	metformin or a sulfonylurea for 3 months	1
Hattori	Overweight	1	Any antidiabetic medications	2
Garvey et al	Nil	0	metformin for at least 10 weeks	1
Osonoi et al	Moderate albuminuria: albumin-to-creatinine ratio of 30.0–299.9 mg/g creatinine	4	Any antidiabetic medications	2
Bouchi et al	Overweight	1	Any antidiabetic medication insulin	2
Tobita et al	NASH One to three of: metabolic syndrome, hypertension and dyslipidaemia	9	Any antidiabetic except insulin	2
Okamoto et al	Overweight	1	Any antidiabetic medication	2
Ferrannini et al	Overweight	1	None	0

Supplementary table 5a – SGLT2i AND Inflammation biomarker Search Strategy

Medline - Inflammation		
1	exp Sodium-Glucose Transporter 2 Inhibitors/	1941
2	gliflozin*.mp.	78
3	(sglt$2 inhibitor* or sglt2 inhibitor* or sodium glucose cotransporter 2 inhibitor*).mp.	1784
4	1 or 2 or 3	2544
5	exp Oxidative Stress/	127771
6	oxidative stress.mp.	185263
7	exp Biomarkers/	716403
8	biomarker*.mp.	486108
9	5 or 6 or 7 or 8	962286
10	4 and 9	236
Embase - Inflammation		
1	exp Sodium-Glucose Transporter 2 Inhibitors/	9226
2	gliflozin*.mp.	181

3	(sglt$2 inhibitor* or sglt2 inhibitor* or sodium glucose cotransporter 2 inhibitor*).mp. [mp=title, abstract, heading word, drug trade name, original title, device manufacturer, drug manufacturer, device trade name, keyword, floating subheading word, candidate term word]	6452
4	1 or 2 or 3	9800
5	exp Oxidative Stress/	276341
6	oxidative stress.mp.	322267
7	exp Biomarkers/	288044
8	biomarker*.mp.	391559
9	5 or 6 or 7 or 8	777768
10	4 and 9	502
Web of Science - Inflammation		
#1	TS=((gliflozin* or sglt$2 inhibitor* or sglt2 inhibitor* or sodium glucose cotransporter 2 inhibitor*) and (oxidative stress or biomarker*)) Indexes=SCI-EXPANDED, SSCI, A&HCI, CPCI-S, CPCI-SSH, ESCI Timespan=All years	208
Cochrane Library - Inflammation		
1	Trials matching ((gliflozin* or sglt$2 inhibitor* or sglt2 inhibitor* or sodium glucose cotransporter 2 inhibitor*) and (oxidative stress or biomarker*)) in Title Abstract Keyword - (Word variations have been searched)	100

Supplementary table 5b – SGLT2i AND Oxidative stress biomarker Search Strategy

Medline – Oxidative stress		
1	exp Sodium-Glucose Transporter 2 Inhibitors/	1981
2	gliflozin*.mp.	90
3	(sglt$2 inhibitor* or sglt2 inhibitor* or sodium glucose cotransporter 2 inhibitor*).mp.	2743
4	1 or 2 or 3	3532
5	exp Inflammation/	323188
6	inflammat*.mp.	996341
7	exp Biomarkers/	719201
8	biomarker*.mp.	543320
9	5 or 6 or 8 or 9	1789936
10	4 and 9	375
Embase - Oxidative stress		
1	exp Sodium-Glucose Transporter 2 Inhibitors/	9320

2	gliflozin*.mp.	184
3	(sglt$2 inhibitor* or sglt2 inhibitor* or sodium glucose cotransporter 2 inhibitor*).mp.	6527
4	1 or 2 or 3	9898
5	exp Inflammation/	3172840
6	inflammat*.mp.	1451823
7	exp Biomarkers/	289495
8	biomarker*.mp.	393435
9	5 or 6 or 8 or 9	4504940
10	4 and 9	1677
Web of Science – Oxidative Stress		
#1	TS=((gliflozin* or sglt$2 inhibitor* or sglt2 inhibitor* or sodium glucose cotransporter 2 inhibitor*) and (inflamma* or biomarker*)) Indexes=SCI-EXPANDED, SSCI, A&HCI, CPCI-S, CPCI-SSH, ESCI Timespan=All years	249
Cochrane Library - Oxidative Stress		
1	Trials matching ((gliflozin* or sglt$2 inhibitor* or sglt2 inhibitor* or sodium glucose cotransporter 2 inhibitor*) and (inflamm* or biomarker*)) in Title Abstract Keyword	123