Effects of Sodium-Glucose Cotransporter 2 Inhibitor on Vascular Endothelial and Diastolic Function in Heart Failure With Preserved Ejection Fraction
— Novel Prospective Cohort Study —

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Background: Pathogenesis of heart failure with preserved ejection fraction (HFpEF) may involve endothelial dysfunction and abnormal vascular structure. Sodium-glucose cotransporter 2 (SGLT2) inhibitors have beneficial cardiovascular effects and may improve vascular function in patients with HFpEF.

Methods and Results: We recruited 184 patients with type 2 diabetes and HFpEF (mean age, 66.0±14.4 years) who were scheduled for treatment with SGLT2 inhibitors, had transthoracic echocardiogram to identify diastolic function, and flow-mediated dilation (FMD) to evaluate endothelial function, and assessed ankle-brachial vascular index (CAVI) and carotid intima-media thickness as indices of vascular function and vascular structure, respectively. Body weight, systolic blood pressure, diastolic blood pressure, triglycerides, remnant lipoprotein cholesterol, fasting plasma glucose, hemoglobin A1c, urinary albumin/creatinine ratio, and insulin resistance (IR) decreased, hematocrit and FMD increased significantly, and CAVI decreased significantly, after 12-week treatment (P<0.05). Short-term SGLT2 inhibitors improved diastolic function, significantly reducing the mitral ratios of septal E/early septal annular tissue Doppler velocity (P=0.003) and lateral E/early lateral e’ (P=0.044). On multiple regression statistically significant associations were seen between mean E/e’ and AFMD, CAVI, and IR.

Conclusions: SGLT2 inhibitors can improve diastolic function in patients with type 2 diabetes, suggesting that current treatment policies for diabetes should be re-examined. Further prospective studies with larger sample sizes could provide mechanistic insights into the benefits of SGLT2 inhibitors.

Key Words: Diastolic function; Echocardiography; Endothelial function; Heart failure with preserved ejection fraction (HFpEF); Sodium-glucose cotransporter 2 (SGLT2) inhibitor
weight loss, as discussed elsewhere. SGLT2 inhibitors may also improve vascular function and vascular structure in patients with HFpEF, but so far this remains unproven.

The concept of DM directly affecting myocardial dysfunction dates back to 1954, when Lundbaek observed that myocardial dysfunction was a common DM-related complication present in two-thirds of elderly DM patients. He subsequently became the first to diagnose a specific form of DM-related cardiomyopathy (DMCMP). Almost 20 years later, Rubler et al provided further evidence that cardiomyopathic dysfunction could indeed result directly from DM and not merely indirectly from concomitant coronary artery disease (CAD). That landmark study reported on post-mortem findings of 4 patients with DM-related nephropathy and HF unrelated to valvular, congenital heart disease, alcoholism or significant artery atherosclerosis. They proposed that these patients had a novel DMCMP caused by myocardial microangiopathy or disturbed myocardial metabolism. The use of the term “cardiomyopathy” to indicate this condition corresponds to the currently used definition of cardiomyopathy. Based on presentation and pathology, the clinical phenotype of this DMCMP resembled dilated cardiomyopathy that was induced by toxic agents or viral myocarditis. When becoming symptomatic, DMCMP with a dilated phenotype presents as HFpEF.

Recently, however, most clinical reports on DMCMP describe a phenotype that differs from a dilated cardiomyopathy. The current shift in perception of DMCMP from a dilated to a restrictive phenotype is matched by a rising awareness that many HF patients present with a normalized left ventricle (LV) and a normal left ventricular ejection fraction (LVEF). Such patients are currently considered to have HFpEF. When becoming symptomatic, DMCMP with a restrictive phenotype presents as HFpEF.

This study investigated whether the SGLT2 inhibitors can improve vascular function and vascular structure and LV diastolic performance in patients with HFpEF.

Methods

Subjects

Patients with DM and HFpEF who participated in the study were generally in good health as determined on medical history, physical examination, screening laboratory tests, urinalysis, and echocardiogram. The main inclusion criteria were (1) age ≥20 years and ≤75 years; (2) diagnosis of type 2 DM ≥3 months prior to screening; and (3) hemoglobin A1c (HbA1c) ≥6.5% (48 mmol/mol) and ≤9.4% (79 mmol/mol) during treatment. Exclusion criteria were: (1) type 1 DM or secondary DM; (2) previous use of SGLT2 inhibitors, glucagon-like peptide-1 agonists, or insulin; (3) previous use of metformin with dosage exceeding 750 mg/day; (4) severe infection, scheduled for surgery, or recent serious trauma; (5) moderate or severe cardiac insufficiency (New York Heart Association [NYHA]) class III or higher; (6) serious liver or renal functional failure (serum creatinine ≥1.3 mg/dL or estimated glomerular filtration rate [eGFR] <45 mL/min/1.73 m²); (7) alcohol dependency or use of illicit drugs; (8) pregnant or breastfeeding, possibly pregnant, or planning to become pregnant in the study period (female patients only); (9) dehydration (abnormal hematocrit [Ht] and blood urea nitrogen, symptoms of dehydration); (10) urinary tract or genital infection; (11) history of hypersensitivity to the study drugs; (12) severe ketosis, diabetic coma or precoma; and (13) otherwise considered unsuitable by the attending physician.

Current diagnostic criteria for HFpEF from the American Society of Echocardiography (ASE)/European Association of Cardiovascular Imaging (EACVI) include clinical signs or symptoms (such as dyspnea, orthopnea, paroxysmal nocturnal dyspnea), EF ≥50%, and evidence of diastolic function. Similar diagnostic criteria for HFpEF from the European Society of Cardiology (ESC) include signs and symptoms, EF ≥50%, and elevated B-type natriuretic peptide (BNP) as well as echocardiographic evidence of diastolic dysfunction or structural abnormalities.

Exact criteria for the grading of diastolic dysfunction have evolved, but diastolic abnormalities on echocardiography are common in community cohorts of patients.

Figure 1. Flowchart of patient selection.
with comorbidities such as obesity, hypertension, DM and CAD, and have poor correlation with HF symptoms. In this study, we evaluated the diagnostic performance of current algorithms by first coding the diagnosis of HFpEF from resting echocardiography data alone, for which we used contemporary diagnostic schemes as proposed by the ESC, and then separately as recommended by the ASE/EACVI to assess diastolic function as follows.

1. Left atrium (LA) volume index >34 mL/m².
2. LV mass index (LVMi) >115 g/cm² (male), >95 g/cm² (female).
3. Mitral early diastolic velocity/early annular tissue Doppler velocity (E/e') >13, means of the e' septal and lateral wall <9 cm/s. (Modified from Oeing et al.)

(A) Algorithm for diagnosis of LV diastolic dysfunction in subjects with normal LVEF.

(B) Algorithm for estimation of LV filling pressures and grading LV diastolic function in patients with depressed LVEF and patients with myocardial disease and normal LVEF, after consideration of clinical and other 2-D data. (Modified from Nagueh et al.)

### Study Protocol

This study involved Japanese patients with HFpEF and type 2 DM who were scheduled to receive treatment with SGLT2 inhibitors as used in clinical practice (empagliflozin 10–25 mg, luseogliflozin 2.5–5 mg, or tofogliflozin 20 mg) at Kenwakai Otemachi Hospital from January 2015 to December 2017. After consecutive screening, eligible patients were enrolled and followed for 12 weeks. Patients were assessed on a regular basis throughout the study.

We investigated multiple early-stage risk factors associated with the development of arteriosclerosis, and we checked the influence of the following risk factors for early onset of acute myocardial infarction, stroke and death: gender, smoking, hypertension, high total cholesterol (TC), high triglycerides (TG), low high-density lipoprotein cholesterol (HDL-C), homeostatic model assessment of insulin resistance (HOMA-IR) index, BNP, urinary albumin, and eGFR. Transthoracic echocardiograms were performed to identify diastolic function. We evaluated endothelial function by measuring flow-mediated dilation (FMD), and we applied cardio-ankle vascular index (CAVI) and carotid

### Table 1. Baseline Patient Characteristics

|                         | All patients (n=184) | Empagliflozin (n=59) | Luseogliflozin (n=63) | Tofogliflozin (n=62) |
|-------------------------|----------------------|----------------------|-----------------------|----------------------|
| Age (years)             | 66.0±14.4            | 62.0±9.4             | 70.3±11.4             | 66.0±9.8             |
| Male (%)                | 60.9                 | 61.5                 | 42.9                  | 78.6                 |
| Body height (cm)        | 161.3±9.4            | 159.4±8.4            | 160.0±9.2             | 166.0±9.5            |
| Body weight (kg)        | 66.1±11.7            | 67.5±10.6            | 55.0±8.7              | 75.0±11.2            |
| BMI (kg/m²)             | 24.6±7.7             | 27.5±7.9             | 21.3±6.5              | 27.8±8.6             |
| SBP (mmHg)              | 139.7±18.8           | 140.7±19.4           | 141.3±21.4            | 137.2±17.8           |
| DBP (mmHg)              | 80.4±13.9            | 79.9±12.8            | 78.4±13.9             | 82.8±13.1            |
| TC (mg/dL)              | 203.4±23.8           | 222.1±22.9           | 188.5±20.8            | 207.3±21.6           |
| LDL-C (mg/dL)           | 105.3±13.8           | 101.0±15.4           | 98.8±10.9             | 115.5±14.5           |
| HDL-C (mg/dL)           | 60.9±11.8            | 62.7±11.9            | 68.3±12.7             | 52.1±10.8            |
| TG (mg/dL)              | 236.3±25.4           | 239.0±61.3           | 226.0±54.2            | 244.0±70.7           |
| L/H                     | 2.01±2.8             | 1.76±2.4             | 1.67±2.1              | 2.29±3.1             |
| Non-HDL (mg/dL)         | 138.7±20.1           | 159.4±25.2           | 115.2±19.3            | 155.2±24.2           |
| RLP-C (mg/dL)           | 15.9±3.8             | 18.9±10.3            | 14.8±1.8              | 13.9±3.5             |
| HbA1c (%)               | 7.2±1.1              | 6.8±1.2              | 7.3±3.1               | 7.6±1.2              |
| FPG (mg/dL)             | 158.7±34.7           | 152.4±30.6           | 151.3±44.7            | 171.4±34.7           |
| IRI (μu/mL)             | 8.60±3.6             | 7.8±2.6              | 2.8±2.5               | 11.6±4.3             |
| HOMA-IR                 | 3.57±1.1             | 2.95±0.9             | 2.85±0.8              | 4.90±1.8             |
| BNP (pg/mL)             | 196.7±31.4           | 179.0±29.3           | 230.2±33.2            | 181.9±30.6           |
| UACR (mg/gCr)           | 89.4±5.4             | 129.7±8.9            | 37.1±3.8              | 98.9±6.8             |
| EF (%)                  | 58±10.8              | 62±12.5              | 55±10.1               | 57±10.5              |
| FS (%)                  | 34±6.3               | 37±7.3               | 32±5.9                | 34±6.1               |
| IVS (mm)                | 11±3.2               | 11±3.7               | 10±3.0                | 11±3.1               |
| LVPW (mm)               | 10±3.1               | 10±3.6               | 9±2.9                 | 10±3.0               |
| E (cm/s)                | 60.4±5.4             | 57.0±4.8             | 59.8±5.1              | 64.4±6.3             |
| A (cm/s)                | 67.6±6.1             | 74.0±7.4             | 67.6±6.4              | 61.3±5.5             |
| E/A                     | 0.89±2.3             | 0.77±2.1             | 0.88±2.6              | 1.05±2.4             |
| Sep e' (cm/s)           | 4.27±1.2             | 4.05±0.8             | 4.26±1.1              | 4.52±1.8             |
| Lat e' (cm/s)           | 4.14±1.6             | 3.89±0.6             | 4.10±0.9              | 4.43±1.5             |
| SepE/e'                 | 14.15±2.0            | 14.07±3.1            | 14.07±3.1             | 14.25±2.0            |
| LatE/e'                 | 14.59±2.1            | 14.65±2.2            | 14.59±2.2             | 14.54±2.1            |
| Mean E/e'               | 14.37±2.2            | 14.36±2.4            | 14.33±1.9             | 14.40±2.8            |
| FMD (%)                 | 4.5±2.2              | 4.6±2.3              | 4.3±2.8               | 4.6±2.1              |

(Table 1 continued the next page.)
All patients  
(n=184)  
Empagliflozin  
(n=59)  
Luseogliflozin  
(n=63)  
Tofogliflozin  
(n=62)

| CAVI |  |  |  |
|---|---|---|---|
| Right | 9.24±2.6 | 8.92±2.1 | 9.33±3.6 | 9.46±2.7 |
| Left | 9.28±1.7 | 8.83±1.4 | 9.77±2.7 | 9.22±1.8 |

| Carotid IMT |  |  |  |
|---|---|---|---|
| Right (mm) | 0.86±0.6 | 0.86±0.7 | 0.84±0.4 | 0.87±0.5 |
| Left (mm) | 0.90±0.2 | 0.95±0.4 | 0.93±0.7 | 0.93±0.3 |

| Ht (%) | 39.5±3.9 | 39.5±4.1 | 36.3±3.1 | 42.8±5.3 |

| History (%) |  |  |  |
|---|---|---|---|
| CAD | 63.2±14.1 | 65.4±14.6 | 62.2±13.9 | 55.7±12.4 |
| Hypertension | 71.8±16.0 | 66.8±14.9 | 85.8±19.1 | 72.6±16.2 |
| Hyperlipidemia | 51.0±11.4 | 49.9±11.1 | 56.4±12.6 | 48.0±10.7 |
| AF | 33.7±7.5 | 32.2±7.2 | 38.6±8.6 | 32.9±7.3 |

| Therapy (%) |  |  |  |
|---|---|---|---|
| ACE or ARB | 73.1±16.3 | 74.2±16.6 | 65.5±14.6 | 79.1±17.6 |
| β-blockers | 85.8±19.1 | 87.4±19.5 | 79.1±17.6 | 88.7±19.8 |
| CCB | 53.8±12.0 | 54.6±12.2 | 48.2±10.8 | 58.2±13.0 |
| Loop diuretics | 71.3±15.9 | 72.1±16.1 | 76.6±17.1 | 61.3±13.7 |
| Thiazide diuretics | 13.2±2.9 | 9.7±2.2 | 21.4±4.8 | 15.4±3.4 |
| Digoxin | 13.4±3.0 | 15.7±3.5 | 8.7±1.9 | 11.3±2.5 |
| Warfarin | 29.6±6.6 | 32.2±7.2 | 25.4±5.7 | 25.6±5.7 |
| Statins | 52.1±11.6 | 52.5±11.7 | 54.3±12.1 | 47.1±10.7 |
| Aspirin | 32.9±7.3 | 35.4±7.9 | 31.4±7.0 | 25.3±5.6 |
| DPP-4 inhibitors | 56.0±12.9 | 58.9±13.1 | 52.0±11.6 | 62.7±14.0 |
| Biguanide | 43.2±9.6 | 46.5±10.4 | 41.2±9.2 | 33.2±7.4 |
| Sulfonylurea | 13.4±3.0 | 15.7±3.5 | 8.7±1.9 | 11.3±2.5 |
| Insulins | 3.4±3.0 | 5.7±3.5 | 4.7±1.9 | 3.3±2.5 |

Data given as mean±SD. HOMA-IR=IRI(μu/mL)×FPG(mg/dL)÷405. A, late diastolic velocity; ACE, angiotensin-converting enzyme; AF, atrial fibrillation; ARB, angiotensin receptor blocker; BMI, body mass index; BNP, brain natriuretic peptide; CAD, coronary artery disease; CAVI, cardio-ankle vascular index; CCB, calcium channel blockers; DBP, diastolic blood pressure; DPP-4, dipeptidyl peptidase 4; E, mitral early diastolic velocity; E/A, mitral early diastolic velocity/late diastolic velocity ratio; E/e', mitral early diastolic velocity/early annular tissue Doppler velocity; EF, ejection fraction; FMD, flow-mediated dilation; FPG, fasting plasma glucose; FS, fractional shortening; HbA1c, hemoglobin A1c; HDL-C, high-density lipoprotein cholesterol; HOMA-IR, homeostatic model assessment of insulin resistance; Ht, hematocrit; IMT, intima-media thickness; IRI, immunoreactive insulin; IVS, interventricular septum; Lat e', early lateral annular tissue Doppler velocity; LatE/e', lateral mitral early diastolic velocity/early lateral annular tissue Doppler velocity; LVPW, left ventricular posterior wall; non-HDL, non-high-density lipoprotein cholesterol; RLP-C, remnant-like particle cholesterol; SBP, systolic blood pressure; Sep e', early septal annular tissue Doppler velocity; SepE/e', septal mitral early diastolic velocity/early septal annular tissue Doppler velocity; TC, total cholesterol; TG, triglyceride; UACR, urinary albumin/creatinine ratio.

intima-media thickness as indices to assess vascular function and vascular structure, respectively.

The study was conducted according to the Declaration of Helsinki. The protocol was reviewed and approved by the institutional review board of each participating center, and all participants provided informed consent.

Endpoints
The primary endpoint of this study was the change in E/e’, defined according to the diagnostic schemes as proposed by the ESC,16 and separately according to recommendations from the ASE/EACVI17 for the assessment of diastolic function.

The secondary endpoints were change in fasting plasma glucose (FPG), HOMA-IR, and FMD.

Measurements
Clinical and biochemistry data were collected at baseline and after the 12-week treatment period. Overnight fasting blood samples were obtained ≤12 weeks after the start of SGLT2 inhibitor (empagliflozin, luseogliflozin, or tofogliflozin) treatment. FMD was conducted at Kenwakai Otemachi Hospital Medical Center. All blood tests were conducted after overnight fasting. Measurement of the following parameters was outsourced to a central laboratory (SRL Laboratory, Tokyo Japan): plasma insulin, BNP, and urinary albumin/creatinine ratio (UACR). Non-HDL-C was estimated by subtracting HDL-C from TC concentration. Remnant-like particle cholesterol (RLP-C) was measured on the direct homogeneous method (MetaboRead, Kyowa Medex, Tokyo, Japan). HOMA-IR was calculated as follows: HOMA-IR=(immunoreactive insulin [μu/mL]×FPG[mg/dL])÷405. Other biochemistry safety parameters (e.g., red cell count, Ht, and uric acid) were also measured.

FMD
The FMD was measured using the UNEX EF38G (UNEX Corporation, Nagoya, Japan) by a technologist who was not a participant in the study and was blinded to the study groups. The methodology has been described in detail previously.20,21 Briefly, all measurements were performed under fasting and non-smoking conditions in the early morning in a temperature-controlled room (25°C). After the patient rested for at least 15 min, the pressure cuff was placed on the forearm to capture baseline images of the brachial artery using high resolution ultrasound. The cuff
was then inflated and kept at 50 mmHg above the systolic BP (SBP) to occlude the brachial artery. The cuff was released 5 min later, and the image of the brachial artery was captured. The diameters of the brachial artery in the pre- and post-hyperemia images were used to calculate changes in FMD according to the following formula: FMD (%)=[(maximum diameter−diameter at rest)×100/diameter at rest].

**Echocardiography**

Comprehensive 2-D, M-mode, conventional Doppler, and tissue Doppler echocardiography was performed according to contemporary guidelines by experienced doctors and sonographers. Mitral early diastolic velocity (E) and late diastolic velocity (A) were measured at the mitral leaflet tips on pulse wave Doppler. Tissue Doppler echocardiography was performed to measure the early annular tissue Doppler velocity (e’) at the ventricular septum and lateral wall. All measures represent the mean of 3 beats for patients in sinus rhythm.

### Table 2. Change in Parameters After 12-Week SGLT2 Inhibitor Treatment

| Parameter                      | All patients (n=184) | Empagliflozin (n=59) | Luseogluflzin (n=63) | Tofogluflzin (n=62) |
|-------------------------------|----------------------|----------------------|----------------------|----------------------|
| Body weight (kg)              | Before               | After                | Before               | After                |
|                               | 66.1±11.7            | 63.0±10.6*           | 67.5±10.6            | 66.7±11.8            |
| Δ                             | -3.1±3.0             | -0.8±0.2             | 55.0±8.7             | 51.7±8.7*            |
|                               | 24.6±7.7             | 24.3±7.5             | 27.5±7.9             | 26.4±7.8             |
| Δ                             | -0.3±0.9             | 11.0±1.0             | 21.3±6.5             | 20.0±5.8*            |
|                               | 139.7±18.8           | 123.5±12.5**         | 140.7±19.4           | 128.9±13.1**         |
| Δ                             | -16.2±8.2            | -11.8±7.0            | 141.3±21.4           | 120.9±14.5**         |
|                               | 80.4±13.9            | 72.2±8.8*            | 79.9±12.8            | 74.1±8.9*            |
| Δ                             | -8.2±4.2             | -5.8±2.4             | 78.4±13.9            | 68.2±10.8*           |
|                               | 203.4±23.8           | 175.3±21.2           | 222.1±22.9           | 190.7±20.8           |
| TC (mg/dL)                    | 207.3±21.6           | 185.7±18.8           | 188.5±20.8           | 158.7±18.8           |
| Δ                             | -28.1±7.6            | -31.4±7.8            | -29.8±7.1            | -26.6±8.8            |
| LDL-C (mg/dL)                 | 15.9±13.9            | 92.8±11.6            | 101.0±15.4           | 104.7±13.1           |
| Δ                             | -12.5±6.4            | +3.7±2.4             | 98.8±10.9            | 78.2±10.1            |
|                               | 60.9±11.8            | 59.9±10.6            | 62.7±11.9            | 57.5±9.7             |
| Δ                             | -1.0±0.10            | -5.2±0.40            | 68.3±12.7            | 65.0±11.5            |
| TG (mg/dL)                    | 236.3±74.7           | 213.8±54.9*          | 239.0±61.3           | 214.6±56.1*          |
| Δ                             | -22.5±7.4            | -24.4±7.6            | 226.0±54.2           | 208.2±34.7*          |
| L/H                           | 2.01±0.8             | 1.67±2.1             | 1.76±2.4             | 1.97±2.8             |
| Δ                             | -0.34±0.14           | 0.21±0.09            | 1.67±2.1             | 1.31±1.8             |
| Non-HDL-C (mg/dL)             | 137.8±20.1           | 115.5±18.4           | 159.4±25.2           | 133.2±20.2           |
| Δ                             | -23.2±7.5            | -26.2±7.8            | 115.2±19.3           | 93.7±18.1            |
| RLP-C (mg/dL)                 | 15.9±3.3             | 8.3±3.3**            | 18.9±10.3            | 8.5±6.3*             |
| Δ                             | -7.0±3.4             | 4.4±7.4              | 14.8±1.8             | 7.9±31*              |
| HbA1c (%)                     | 7.2±1.1              | 6.7±0.7*             | 6.8±1.2              | 6.5±1.2              |
| Δ                             | -0.50±0.9            | -0.3±0.66            | 7.3±3.1              | 6.7±2.7              |
| FPG (mg/dL)                   | 158.7±34.7           | 134.1±19.0**         | 152.4±30.6           | 136.4±19.3**         |
| Δ                             | -24.6±3.4            | -16.0±8.4            | 151.3±44.7           | 132.1±39.0**         |
| IRI (µu/mL)                   | 8.60±3.6             | 8.2±3.1              | 7.8±2.6              | 7.5±2.4              |
| Δ                             | -0.36±0.16           | -0.27±0.12           | 7.6±2.5              | 7.45±2.3             |
| HOMA-IR                       | 3.57±1.1             | 2.73±0.8*            | 2.95±0.9             | 2.55±0.7*            |
| Δ                             | -0.84±0.17           | -0.40±0.06           | 2.85±0.8             | 2.43±0.6*            |
| BNP (pg/mL)                   | 196.7±31.4           | 203.7±33.5           | 179.0±29.3           | 126.4±22.4           |
| Δ                             | +7.0±3.1             | -52.6±8.1           | 230.2±33.2           | 200.1±31.8           |
| UACR (mg/gCr)                 | 89.4±5.4             | 47.4±3.1**           | 129.7±8.9            | 95.9±6.7**           |
| Δ                             | -42.0±4.8            | -33.8±4.1           | 37.1±3.8             | 15.3±2.1**           |

Statistical Analysis

Data are expressed as mean±SD, number and percentage, or percent change after treatment. Statistical analyses were performed using SAS version 9.3 (SAS Institute, Cary, NC, USA). The unpaired t-test (for continuous variables) or Fisher’s exact test (for categorical variables) was used for statistical analysis of differences in the baseline clinical parameters in all patients and in all 3 subgroups (empagliflozin, luseogliflozin, and tofogliflozin). Plasma parameters before and after treatment were compared on paired t-test. For intergroup comparisons in the subgroups, unpaired t-test was used for normally distributed data and the Mann-Whitney U-test, for data with skewed distribution. To determine correlations between 2 variables, the Pearson correlation coefficient was used for data with a normal distribution pattern and the Spearman rank-correlation coefficient, for data with non-normal distribution. Stepwise multiple regression analysis was carried out using the changes in mean E/e’ from baseline in week 12 of treatment as the dependent variable and age, body mass index, HbA1c, FMD,
CAVI, and HOMA-IR as independent variables. Differences were considered statistically significant at P<0.05.

Results

We assessed 190 patients with DM and HFpEF, and we enrolled 184. We excluded 6 patients whose condition deteriorated suddenly into HFpEF (EF <40%) or who had recently been diagnosed with severe valvular disease or amyloidosis. The ratio of patients who were treated with each of the 3 SGLT2 inhibitors (empagliflozin, luseogliflozin, or tofogliflozin) was approximately 1:1:1. All enrolled patients completed the study (Figure 1).

Mean age at baseline was 66.0±14.4 years; 60.9% of patients were male. In all patients, BP was relatively well controlled by antihypertensive drugs, and low-density lipoprotein cholesterol (LDL-C) was well managed with statins. Baseline laboratory data, however, were high for TG and RLP-C, and also for FPG and HbA1c (not well-controlled). Baseline characteristics were similar across the 3 groups of patients (Table 1).

After 12 weeks of SGLT2 inhibitor treatment, we noted significant reductions in body weight (BW; mean±SD, −3.1±3.0 kg, P<0.05), SBP (−16.2±8.2 mmHg, P<0.01), diastolic BP (DBP; −8.2±4.2 mmHg, P<0.01), TG (−22.5±7.4 mg/dL, P<0.01), RLP-C (−7.6±3.4 mg/dL, P<0.01), HbA1c (−0.5±0.9%, P<0.05), FPG (−24.6±3.4 mg/dL, P<0.01), UACR (−42.0±48.0, P<0.01), and HOMA-IR (−0.8±0.17, P<0.05) in the study group. Ht increased significantly after 12 weeks of treatment (2.3±1.4%, P<0.05). FMD was significantly higher and CAVI was significantly lower (2.6±1.1%, P<0.05; −0.6±0.29, P<0.05; Table 2; Figure 2). We noted significant reductions in E/A ratio, (mean±SD, −0.6±0.007; P=0.048); septal E’/e’ (−6.7±3.0; P=0.003), and lateral E’/e’ (−8.3±0.008, P=0.044; Table 3; Figure 2).

Changes of outcome in the subgroups were similar to the changes seen in the total group (Tables 2.3). There were no statistically significant differences in outcome between the 3 groups in this class.

| Table 1 |

|                        | All patients (n=184) | Empagliflozin (n=59) | Luseogliflozin (n=63) | Tofogliflozin (n=62) |
|------------------------|----------------------|----------------------|-----------------------|----------------------|
| E (cm/s)               | 60.4±5.4             | 57.3±4.9             | 55.4±4.3              | 57.4±5.1             |
| Δ                      | −3.1±2.2             | −1.6±1.8             | −2.4±2.0              | −5.4±3.1             |
| A (cm/s)               | 67.6±6.1             | 71.2±6.9             | 70.0±6.7              | 61.3±5.5             |
| Δ                      | +1.2±1.1             | −2.8±2.6             | +2.4±2.1              | −4.0±2.9             |
| E/A                    | 0.89±2.3             | 0.78±1.8             | 0.82±2.5              | 1.05±2.4             |
| Δ                      | −0.06±0.007          | +0.01±0.002          | −0.06±0.007           | −0.15±0.011          |
| Sep e’ (cm/s)          | 4.27±1.2             | 4.05±0.8             | 4.25±1.1              | 4.52±1.8             |
| Δ                      | +3.45±0.004          | +4.34±0.005          | +2.68±0.001           | +3.34±0.003          |
| Lat e’ (cm/s)          | 4.14±1.6             | 3.89±0.6             | 4.10±0.9              | 4.43±1.5             |
| Δ                      | +5.05±0.007          | +4.97±0.006          | +4.62±0.006           | +5.55±0.009          |
| SepE/e’                | 14.15±2.0            | 14.07±3.1            | 14.07±3.1             | 14.25±2.0            |
| Δ                      | −6.73±0.009          | −7.47±0.011          | −5.76±0.007           | −6.74±0.010          |
| LatE/e’                | 14.59±2.1            | 14.65±2.2            | 14.59±2.2             | 14.54±2.1            |
| Δ                      | −8.35±0.008          | −8.40±0.008          | −8.01±0.006           | −8.63±0.009          |
| Mean E/e’              | 14.37±2.2            | 14.36±2.4            | 14.33±1.9             | 14.40±2.8            |
| Δ                      | −7.54±0.007          | −7.93±0.008          | −6.88±0.006           | −7.17±0.008          |
| FMD (%)                | 4.57±2.2             | 4.62±2.3             | 4.32±2.8              | 4.62±2.1             |
| Δ                      | 2.66±1.19            | 2.90±1.21            | 2.60±1.18             | 2.50±1.16            |
| CAVI (Right)           | 9.24±2.6             | 8.92±2.1             | 9.33±3.6              | 9.46±2.7             |
| Δ                      | −0.74±0.31           | −0.09±0.05           | −0.96±0.41            | −1.16±0.51           |
| CAVI (Left)            | 9.28±1.7             | 8.73±1.1             | 9.22±1.8              | 8.28±2.1             |
| Δ                      | −0.57±0.25           | −0.10±0.06           | −0.65±0.29            | −0.94±0.29           |
| Mean CAVI              | 9.26±2.3             | 8.78±2.2             | 9.55±2.6              | 9.34±2.4             |
| Δ                      | −0.65±0.29           | −0.10±0.06           | −0.80±0.32            | −1.05±0.48           |
| Carotid IMT (Right) (mm)| 0.86±0.6             | 0.86±0.7             | 0.84±0.7              | 0.87±0.5             |
| Δ                      | −0.04±0.01           | −0.08±0.04           | 0.10±0.07             | −0.14±0.08           |
| Carotid IMT (Left) (mm)| 0.92±0.2             | 0.95±0.4             | 0.93±0.3              | 0.93±0.3             |
| Δ                      | −0.09±0.05           | −0.25±0.12           | ±0.00±0.02            | −0.15±0.09           |
| Mean carotid IMT (mm)  | 0.88±0.2             | 0.91±0.1             | 0.84±0.2              | 0.90±0.1             |
| Δ                      | −0.05±0.02           | −0.13±0.08           | 0.10±0.06             | −0.14±0.09           |
| Ht (%)                 | 39.5±3.9             | 41.8±4.2             | 36.3±3.1              | 42.8±5.3             |
| Δ                      | 2.3±1.7              | 4.3±3.1              | 2.1±1.6              | 0.6±0.3              |

Data given as mean±SD. *P<0.05, **P<0.01 vs. baseline. HOMA-IR=IRI(μu/mL)×FPG(mg/dL)+405. SGLT2, sodium-glucose cotransporter 2. Other abbreviations as in Table 1.
Figure 2. Changes in markers before and after 12-week sodium-glucose cotransporter 2 (SGLT2) inhibitor treatment. Bars, mean±SD. *P<0.05, **P<0.01 vs. baseline. IR was calculated using the formula HOMA-IR=IRI(μu/mL)×FPG(mg/dL)÷405. CAVI, cardio-ankle vascular index (L, left; R, right); FMD, flow-mediated dilation; FPG, fasting plasma glucose; HOMA-IR, homeostatic model assessment of insulin resistance; IR, insulin resistance; IRI, immunoreactive insulin; LatE/e’, lateral mitral early diastolic velocity/early lateral annular tissue Doppler velocity; RLP-C, remnant-like particle cholesterol; SepE/e’, septal mitral early diastolic velocity/early septal annular tissue Doppler velocity.

Table 3. Marker Levels Before and After 12-Week SGLT2 Inhibitor Treatment

|                      | All patients (n=184) | Empagliflozin (n=59) | Luseogliflozin (n=63) | Tofogliflozin (n=62) |
|----------------------|----------------------|-----------------------|-----------------------|-----------------------|
|                      | Before               | After                 | Before               | After                 | Before               | After                 | Before               | After                 |
| SBP (mmHg)           | 139.7±18.8           | 123.5±12.5*           | 140.7±19.4           | 128.9±13.1**          | 141.3±21.4           | 120.9±14.5**          | 137.2±17.8           | 120.8±12.5*          |
| DBP (mmHg)           | 80.4±13.9            | 72.2±8.8**            | 79.9±12.8            | 74.1±8.9*             | 78.4±13.9            | 68.2±10.8**           | 82.8±13.1            | 74.6±8.6*            |
| FMD (%)              | 4.50±2.2             | 7.16±3.2*             | 4.6±2.3              | 7.3±3.1*              | 4.3±2.8              | 6.9±3.3*              | 4.6±2.1              | 7.1±3.4*             |
| CAVI                 |                      |                       |                       |                       |                       |                       |                       |                       |
| Right                | 9.24±2.6             | 8.50±2.1*             | 8.92±2.1             | 8.83±2.2*             | 9.33±3.6             | 8.37±3.3*             | 9.46±2.7             | 8.30±2.3*            |
| Left                 | 9.28±1.7             | 8.71±1.3*             | 8.83±1.4             | 8.73±1.1*             | 9.77±2.7             | 9.12±3.3*             | 9.22±1.8             | 8.28±2.1*            |
| Mean CAVI            | 9.26±2.3             | 8.61±1.9              | 8.88±1.8             | 8.78±2.2              | 9.55±2.6             | 8.75±2.8              | 9.34±2.4             | 8.29±2.1             |
| Carotid IMT (mm)     |                      |                       |                       |                       |                       |                       |                       |                       |
| Right                | 0.86±0.6             | 0.82±0.3              | 0.86±0.7             | 0.78±0.2              | 0.84±0.7             | 0.94±0.4              | 0.87±0.5             | 0.73±0.3             |
| Left                 | 0.92±0.2             | 0.83±0.5              | 0.95±0.4             | 0.77±0.4              | 0.93±0.3             | 0.93±0.7              | 0.93±0.3             | 0.78±0.6             |
| Mean IMT (mm)        | 0.88±0.2             | 0.83±0.1              | 0.91±0.1             | 0.78±0.1              | 0.84±0.2             | 0.94±0.3              | 0.90±0.1             | 0.76±0.1             |
| SepE/e’              | 14.15±2.0            | 7.42±1.2**            | 14.07±3.1            | 6.6±1.4**             | 14.07±3.1            | 8.31±3.0**            | 14.25±2.0            | 7.51±1.6**           |
| LatE/e’              | 14.59±2.1            | 6.24±1.7**            | 14.65±2.2            | 6.25±2.0**            | 14.59±2.2            | 6.58±3.7**            | 14.54±2.1            | 5.91±1.1**           |
| Mean E/e’            | 14.37±2.2            | 6.83±1.8              | 14.36±2.4            | 6.43±1.2**            | 14.33±1.9            | 7.45±2.0**            | 14.40±2.8            | 7.23±1.9**           |
| FPG (mg/dL)          | 158.7±34.7           | 134.1±19.0**          | 152.4±30.6           | 136.4±19.3**          | 151.3±44.7           | 132.1±39.0**          | 171.4±34.7           | 132.8±41.0**         |
| HbA1c (%)            | 7.2±1.1              | 6.7±0.7*              | 6.8±1.2              | 6.5±1.2*              | 7.3±3.1              | 6.7±2.7*              | 7.6±1.2              | 6.9±0.5*             |
| TG (mg/dL)           | 236.3±74.7           | 213.6±54.9*           | 239.0±61.3           | 214.6±56.1*           | 226.0±54.2           | 208.2±34.7*           | 244.0±70.7           | 216.8±55.8*          |
| RLP-C (mg/dL)        | 15.9±3.3             | 8.3±3.3*              | 18.9±10.3            | 8.5±6.3*              | 14.8±1.8             | 7.9±313*              | 13.9±3.5             | 8.5±5.4*             |
| HOMA-IR              | 3.57±1.1             | 2.73±0.8*             | 2.95±0.9             | 2.55±0.7*             | 2.85±0.8             | 2.43±0.6*             | 4.90±1.8             | 3.20±1.7*            |

Data given as mean±SD. *P<0.05, **P<0.01 vs. baseline. HOMA-IR=IRI(μu/mL)×FPG(mg/dL)÷405. Abbreviations as in Tables 1,2.
SGLT2 Inhibitors for Heart Function in HFpEF

On multiple regression analysis a statistically significant association was seen between Δmean E/e’ and ΔFMD, ΔCAVI, and ΔHOMA-IR. Interestingly, short-term SGLT2 inhibitors improved diastolic function (Table 4). We found that mean E/e’ in week 12 was influenced significantly by FMD, CAVI, and HOMA-IR, respectively (Table 5).

### Discussion

SGLT2 inhibitors cause excess glucose to be excreted into the urine and provide an anti-hyperglycemic effect. Hyperglycemia has been recognized as a primary factor in endothelial dysfunction, leading to the development of vascular complications and vascular inflammation in diabetic patients. This study showed that SGLT2 inhibitors reduced FPG and prevented the development of endothelial dysfunction and vascular inflammation, at least partially, through a reduction of oxidative stress. Accumulating evidence suggests cardioprotective effects of SGLT2 inhibitors. The present study may elucidate a mechanism for these effects.

Different mechanisms drive LV remodeling in HFpEF and HFrEF. In HFpEF, coronary microvascular endothelial dysfunction drives LV remodeling and dysfunction by lowering myocardial nitric oxide (NO) bioavailability and protein kinase G (PKG) activity. In HFrEF, cardiomyocyte cell death drives LV remodeling and dysfunction, and is caused by oxidative stress in the cardiomyocyte compartment. On histopathology, collagen is laid down between cardiomyocytes and the sarcomeric structure is preserved in HFpEF. In HFrEF, however, the collagen is laid down over extended areas (replacement fibrosis), and the sarcomeric structure disappears. Coronary microvascular endothelial dysfunction is reported to affect HFpEF.

The cardiovascular consequences of DM have been shown to be associated with oxidative stress. Hyperglycemia encourages the non-enzymatic glycosylation of proteins and subsequent formation of advanced glycation end products (AGE), which interact with the receptors for AGE (RAGE) on the plasma membrane and promote the production of reactive oxygen species (ROS). This contributes to vascular complications. Hyperglycemia can also activate polyol pathways and protein kinase C, further promoting ROS production. In this study, 12-week SGLT2 inhibitor treatment resulted in significant reductions in BW, SBP, DBP, TG, RLP-C, FPG, HbA1c, UACR and HOMA-IR. FMD was significantly higher at the end of the 12-week period, and CAVI was significantly lower (P<0.05). We noted a significant reduction in mitral early E/A ratio, septal E/e’, and lateral E/e’e; and on multiple regression statistically significant associations were seen between Δmean E/e’ and ΔFMD, ΔCAVI, and Δ HOMA-IR.

### Table 4. ΔMean E/e’ and ΔFMD, ΔCAVI, ΔIR Correlations

| Variables       | ΔMean E/e’ | ΔFMD | ΔCAVI | ΔHOMA-IR |
|-----------------|------------|------|-------|----------|
| ΔMean E/e’      | r²         | P-value | r²   | P-value  | r²   | P-value |
| ΔFMD            | -0.463     | <0.05* | 0.501 | <0.05*   | 0.446 | <0.05* |
| ΔCAVI           | 0.428      | <0.05* | -0.441| <0.05*   |       |        |
| ΔHOMA-IR        | 0.857      | <0.01**|       |          |       |        |

*P<0.05, **P<0.01. †Coefficient of correlation. HOMA-IR=IRI(μu/mL)×FPG(mg/dL)/405. Abbreviations as in Table 1.

### Table 5. Multivariate Indicators of Change in Mean E/e’ After 3 Months

| Variables | Standardized coefficient | P-value | 95% CI |
|-----------|--------------------------|---------|-------|
| Age       | 0.165                    | 0.732   | -0.290 to 0.041 |
| BMI       | 0.080                    | 0.121   | -0.471 to 0.299 |
| HbA1c     | 0.029                    | 0.463   | -0.405 to 0.508 |
| FMD       | -0.585                   | 0.005** | -0.585 to -0.485 |
| CAVI      | 0.392                    | 0.016*  | 0.048 to 0.456 |
| HOMA-IR   | 0.431                    | 0.034*  | 0.001 to 1.018 |

*P<0.05, **P<0.01. HOMA-IR=IRI(μu/mL)×FPG(mg/dL)/405. Abbreviations as in Table 1.

Overall, SGLT2 inhibitors can improve diastolic function in patients with type 2 DM. This may be because improved insulin resistance is associated with increased NO production, and tetrodorphorylation is stimulated through a cyclic guanosine monophosphate (cGMP)/PKG-dependent pathway. Stimulation of PKG requires cGMP, which is either synthesized by soluble guanylate cyclase (sGC) activated by NO or by receptor guanylate cyclase linked to the nitric peptide receptor. This is, in turn, counterbalanced by hydrolysis of cGMP back to cMP by select members of the phosphodiesterase superfamily. Their inhibition, which leads to increased cGMP, can also increase PKG activity. This mutual increase in activity means that HFrEF and renal dysfunction promote each other. HFrEF promotes renal dysfunction through (1) elevated central venous pressure due to pulmonary hypertension and right ventricular (RV) function, (2) inability to increase cardiac output following arterial vasodilation due to chronotropic incompetence and fixed LV stroke volume, and (3) systemic inflammation, endothelial dysfunction, and low NO bioavailability, which reduces renal blood flow, and sodium excretion. In turn, HFrEF is promoted by worsening systemic inflammation, endothelial dysfunction, and NO bioavailability, due in part to renal-specific mediators. In addition to systemic hemodynamic effects, SGLT2 inhibitors affect renal function directly. After the initial dip, however, eGFR subsequently tends to return toward baseline and remains stable over time. The impact of SGLT2 inhibition on eGFR is consistent in patients with established cardiovascular disease, either with or without chronic kidney disease, and is observed after 3–4 weeks of treatment. In the present study, the eGFR dip was reversed ≤2 weeks after drug discontinuation, and an effect also observed in the EMPA-REG OUTCOME trial. The present study also showed a significant reduction in UACR.
stimulate erythropoiesis and oxygen transport to tissue, and thus play a protective role against cardiovascular disease. The present study also confirmed a significant increase in Ht in patients with type 2 DM after SGLT2 inhibitor treatment. Interestingly, on covariance analysis there was also a significant difference in ΔFMD after accounting for the change in Ht. This suggests that increased Ht might play a role in increased FMD, at least in part due to the SGLT2 inhibitors. The increase in Ht does not seem to be linked to hypovolemia or dehydration, but appears to be associated with normalization of hyperfiltration leading to improved renal function.

Aside from the effects of SGLT2 inhibitors described here, it has been suggested that BP management or weight loss in response to SGLT2 inhibition may mediate changes in kidney function. In type 2 DM, interstitial hypoxia can cause increased glucose absorption in the tubular cells and increased afferent renal neural activity. This enhances sympathetic nerve activity and reduces the baroreceptor reflex response through negative feedback, which in turn leads to fluid retention, vasoconstriction, and increased heart rate. SGLT2 inhibitors may suppress this pathway.

Several clinical studies have shown that SGLT2 inhibitors decrease plasma TG and increase HDL-C, but also increase LDL-C, but in the present study, reductions were seen in both LDL-C and HDL-C concentration, and this concern has also been refuted by the large scale EMPA-REG trial. In addition, recent data from an animal model has demonstrated that SGLT2 inhibitors tend to shift metabolism from carbohydrate toward lipid utilization, which increases ketogenesis and LDL-C concentration despite net lipid metabolic utilization. Some confusion still exists in this area, however, and finding research into this metabolic shift is needed.

BNP stimulates receptor GC to produce cGMP and activate PKG. NO produced by NO synthases stimulates sGC to produce cGMP, which activates PKG. In this study, we evaluated BNP and found no significant difference after treatment with SGLT2 inhibitors.

No statistically significant differences in outcome were seen in the between 3 SGLT2 inhibitors in the present study. The presence of a class effect across the SGLT2 inhibitors remains unclear, because such class effects were not the primary focus of our study. Further investigation is required to clarify this point.

Study Limitations
Although the effectiveness of SGLT2 inhibitors has been demonstrated clearly in the present study, the study has several limitations. First, the number of patients was relatively small, and the duration of the study was relatively short; longer trials with a larger sample size, preferably involving patients of different ethnicities, are clearly needed. Second, patients enrolled in this study had moderate hyperglycemia, therefore the effects of treatment in patients with HbA1c >8% remain unknown.

Conclusions
SGLT2 inhibitors can improve diastolic function in patients with type 2 DM, suggesting that current treatment policies for DM could be re-examined. Further prospective studies with larger sample sizes could provide insights into the mechanisms underlying the benefits of SGLT2 inhibitors.
P, et al. Classification of the cardiomyopathies: A position statement from the European Society of Cardiology Working Group on Myocardial and Pericardial Diseases. *Circulation Reports* Vol.1, July 2019; 29: 270–276.

Forbes JM, Cooper ME. Mechanisms of diabetic complications. *Physiol Rev* 2013; 93: 137–188.

Naglie SG, Smith OA, Appleton CP, Byrd BF 3rd, Dukainish H, Edvardsen T, et al. Recommendations for the evaluation of left ventricular diastolic function by echocardiography: An update from the American Society of Echocardiography and the European Association of Cardiovascular Imaging. *J Am Soc Echocardiogr* 2016; 29: 277–314.

Oelze M, Tschöpe C, Pieske B. The new ESC Guidelines for acute and chronic heart failure 2016. *Hert* 2016; 41: 655–663.

Redfield MM, Jacobsen SJ, Burnett JC Jr, Mahoney DW, Bailey KR, Rodeheffer RJ. Burden of systolic and diastolic ventricular dysfunction in the community. *JAMA* 2003; 289: 194–202.

Abhayaratna WP. Characteristics of left ventricular diastolic dysfunction in the community: An echocardiographic survey. *Heart* 2006; 92: 1259–1264.

Corretti MC, Anderson TJ, Benjamin EJ, Celermajer D, Charbonneau F, Creager MA, et al. Guidelines for the ultrasound assessment of endothelial-dependent flow-mediated vasodilation of the brachial artery: A report of the International Brachial Artery Reactivity Task Force. *J Am Coll Cardiol* 2002; 39: 257–265.

Shigiyama F, Kumashiro N, Miyagi M, Iga R, Kobayashi Y, Kanda E, et al. Linagliptin improves endothelial function in patients with type 2 diabetes: A randomized study of linagliptin effectiveness on endothelial function. *J Diabetes Investig* 2017; 8: 330–340.

Tahara A, Kurosa 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. *Eur J Pharmacol* 2013; 715: 246–255.

Oelze M, Kröller-Schön S, Welschof P, Jansen T, Hausding M, et al. Empagliflozin improves glycemic parameters and cardiovascular function in the community: An echocardiographic survey. *Heart* 2006; 92: 1259–1264.

Corretti MC, Anderson TJ, Benjamin EJ, Celermajer D, Charbonneau F, Creager MA, et al. Guidelines for the ultrasound assessment of endothelial-dependent flow-mediated vasodilation of the brachial artery: A report of the International Brachial Artery Reactivity Task Force. *J Am Coll Cardiol* 2002; 39: 257–265.

Shigiyama F, Kumashiro N, Miyagi M, Iga R, Kobayashi Y, Kanda E, et al. Linagliptin improves endothelial function in patients with type 2 diabetes: A randomized study of linagliptin effectiveness on endothelial function. *J Diabetes Investig* 2017; 8: 330–340.

Tahara A, Kurosa 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. *Eur J Pharmacol* 2013; 715: 246–255.

Oelze M, Kröller-Schön S, Welschof P, Jansen T, Hausding M, et al. The sodium-glucose co-transporter 2 inhibitor empagliflozin improves diabetes-induced vascular dysfunction in the streptozotocin diabetes rat model by interfering with oxidative stress and glucotoxicity. *PLOS ONE* 2014; 9: e112394.

Wu JH, Foote C, Blomster J, Toyama T, Perkovic V, O'Connor PM, Ohsaki Y, Musters RJ, Kupreishvili K, et al. Diastolic stiffness of the failing heart: Importance of fibrosis, advanced glycation end products of medullary flow responses to altered NO availability in humans. *Am J Physiol Regul Integr Comp Physiol* 2010; 299: R1449–R1455.

Cefalu WT, Leiter LA, Xie L, et al. Efficacy and safety of empagliflozin added to existing antidiabetes treatment in patients with type 2 diabetes inadequately controlled with metformin (CANTATA-SU): 52 week results from a randomised, double-blind, placebo-controlled trial. *Lancet Diabetes Endocrinol* 2014; 2: 369–384.

Yale JF, Bakris G, Cariou B, Yue D, David-Neto E, Xi L, et al. Efficacy and safety of canagliflozin in subjects with type 2 diabetes and chronic kidney disease: A randomized, double-blind, placebo-controlled trial. *Lancet Diabetes Endocrinol* 2014; 2: 369–384.

Kohan DE, Fioretto P, Wang T, List JF. Long-term study of patients with type 2 diabetes and moderate renal impairment shows that dapagliflozin reduces weight and blood pressure but does not improve glycemic control. *Kidney Int* 2014; 85: 962–971.

Valdes WT, Leiter LA, Yoon KH, Arias P, Niskannen L, Xie J, et al. Efficacy and safety of canagliflozin versus glimepiride in patients with type 2 diabetes and chronic kidney disease: The CANVAS trial. *Am J Physiol Renal Physiol* 2014; 306: F651–F670.

Ferrannini E, Mark M, Mayoux E. CV protection in the EMPA-REG OUTCOME trial: A “thrifty substrate” hypothesis. *Diabetes Care* 2016; 39: 1108–1114.

Sano M. Hemodynamic effects of sodium-glucose cotransporter 2 inhibitors. *J Clin Med Res* 2017; 9: 457–460.

Ptaszynska A, Hardy E, Johnsson E, Parikh S, List JF. Effects of dapagliflozin on cardiovascular risk factors. *Postgrad Med* 2013; 125: 181–189.

Hach T, Gerich J, Salsai A, Kim G, Hantel S, Woerle HJ, et al. Empagliflozin improves glycemic parameters and cardiovascular risk factors in patients with type 2 diabetes (T2DM): Pooled data from four pivotal phase III trials. *Diabetol Stoffw* 2014; 9: 142 (in German).

Briand F, Mayoux E, Brousseau E, Aulier C, Urban I, Costard C, et al. Empagliflozin, via switching metabolism toward lipid utilization, moderately increases LDL cholesterol levels through reduced LDL catabolism. *Diabetes* 2016; 65: 2032–2038.