Serum high-molecular-weight adiponectin and response to dapagliflozin in patients with type 2 diabetes and non-alcoholic fatty liver disease

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ABSTRACT

A better baseline renal function is associated with a better response to sodium-glucose co-transporter-2 inhibitors in patients with type 2 diabetes. Low serum adiponectin is associated with visceral fat accumulation and hepatic steatosis. We investigated the relationship between baseline serum adiponectin and glycemic response to dapagliflozin in patients with type 2 diabetes and non-alcoholic fatty liver disease (NAFLD). In a randomized, active-controlled, open-label trial, 57 patients with type 2 diabetes and NAFLD were randomized to either the dapagliflozin (5 mg/d) group or the control group. Both groups were treated for 24 weeks. Serum high-molecular-weight (HMW) adiponectin was measured with an ELISA kit. Visceral fat area (VFA) was measured by dual bioelectrical impedance analysis. Hepatic steatosis was assessed by the controlled attenuation parameter (CAP) measured by a transient elastography (FibroScan). Treatment with dapagliflozin significantly decreased HbA1c from 8.4%±1.5% at baseline to 7.4%±1.2% at 24 weeks. Both VFA and CAP decreased in the dapagliflozin group. Baseline serum HMW adiponectin was negatively correlated with changes in HbA1c from baseline to 24 weeks with dapagliflozin therapy. In the multivariate analysis, baseline HbA1c (β=−0.559, p=0.002) and serum HMW adiponectin (β=0.471, p=0.010) were independent determinants for the change (reduction) in HbA1c. In the dapagliflozin group, the change in HbA1c was positively correlated with the changes of CAP, but negatively correlated with the change in serum HMW adiponectin. In conclusion, a lower serum level of HMW adiponectin was associated with a better response to dapagliflozin in patients with type 2 diabetes and NAFLD.

Trial registration number UMIN000022155.

Significance of this study

What is already known about this subject?

- Sodium-glucose co-transporter-2 (SGLT2) inhibitors are the latest class of antidiabetic medication that inhibit the absorption of glucose from the proximal tubule of the kidney and hence cause glycosuria.
- A better baseline renal function is associated with a better response to SGLT2 inhibitors in patients with type 2 diabetes.
- Low serum adiponectin is associated with visceral fat accumulation and hepatic steatosis in type 2 diabetes. SGLT2 inhibitor increases serum levels of adiponectin.

What are the new findings?

- In this study, we investigated whether the baseline serum high-molecular-weight (HMW) adiponectin level was associated with the response to treatment with dapagliflozin for 24 weeks in patients with type 2 diabetes and non-alcoholic fatty liver disease (NAFLD).
- A lower serum level of HMW adiponectin at baseline was associated with a better response to dapagliflozin in patients with type 2 diabetes and NAFLD.

How might these results change the focus of research or clinical practice?

- Clinicians will decide to initiate an administration of SGLT2 inhibitor based on baseline serum HMW adiponectin for people with type 2 diabetes and NAFLD.

INTRODUCTION

Sodium-glucose co-transporter-2 (SGLT2) inhibitors reduce plasma glucose concentrations by increasing urinary glucose excretion.[1 2] This glucosuria results in loss of energy and water via osmotic diuresis and causes a reduction in body weight and body fat.[1 2] Several studies have reported that a higher baseline glycated hemoglobin (HbA1c) and glomerular filtration rate (GFR) are associated with a better response to SGLT2 inhibitors in patients with type 2 diabetes. In contrast, baseline body mass index (BMI) did not influence the glucose-lowering effects of SGLT2 inhibitors.[3–6] We recently reported that the SGLT2 inhibitor dapagliflozin decreases hepatic steatosis and visceral fat mass in patients with type 2 diabetes and non-alcoholic fatty liver disease (NAFLD), and also attenuates liver fibrosis in patients with significant fibrosis,[7] suggesting that SGLT2 inhibitors

Table 1 Univariate and multivariate analyses of relationships between serum high-molecular-weight (HMW) adiponectin and clinical parameters at baseline in a total of 57 patients with type 2 diabetes and NAFLD

	Univariate analysis		Multivariate analysis	
Variable	r	P value	β	P value
Age (y)	0.1491	0.2728	NE	
Body weight (kg)	−0.3352	0.0124	−0.046	0.779
BMI	−0.2313	0.0535	NE	
Body water (kg)	−0.4178	0.0019	0.022	0.893
ECW/TBW	0.4671	0.0004	0.349	0.027
Body fat (kg)	−0.0457	0.7429	NE	
VFA (cm^2)	−0.3383	0.0122	−0.059	0.720
FPG (mg/dL)	0.08436	0.5365	NE	
HbA1c (%)	0.07541	0.5807	NE	
HOMA-IR	−0.1193	0.3857	NE	
Fasting C peptide (ng/mL)	−0.3817	0.0037	−0.070	0.668
LDL cholesterol (mg/dL)	−0.03209	0.8144	NE	
Triglyceride (mg/dL)	−0.3424	0.0098	−0.033	0842
HDL cholesterol (mg/dL)	0.5661	<0.0001	0.365	0.024
eGFR (mL/min/1.73 m^2)	0.07755	0.5700	NE	
Uric acid (mg/dL)	−0.1982	0.1507	NE	
AST (U/L)	−0.2775	0.0384	NE	
ALT (U/L)	−0.4219	0.0012	−0.042	0.795
GGT (U/L)	−0.386	0.0033	−0.110	0.500
Hematocrit (%)	−0.2901	0.0301	0.089	0.586
Serum hs-CRP	−0.2631	0.0501	NE	
Serum type 4 collagen 7S	−0.08232	0.5464	NE	
CAP	−0.313	0.0225	0.004	0.982
R^2			0.454	

β indicates partial coefficient.
ALT, alanine transaminase; AST, aspartate aminotransferase; BMI, body mass index; CAP, controlled attenuation parameter; ECW, extracellular water; eGFR, estimated glomerular filtration rate; FPG, fasting plasma glucose; GGT, γ-glutamyltranspeptidase; HDL, high-density lipoprotein; HOMA-IR, homeostasis model assessment of insulin resistance; hs-CRP, high-sensitivity C-reactive protein; LDL, low-density lipoprotein; NAFLD, non-alcoholic fatty liver disease; NE, does not enter the final model; TBW, total body water; VFA, visceral fat area.

can exert a glucose-lowering effect by improving insulin sensitivity in the liver.

Adiponectin is an adipocyte-specific protein that enhances insulin sensitivity, promotes lipid metabolism, and has an anti-inflammatory or antiatherogenic effect.[8] Serum adiponectin levels are lower in people with metabolic syndrome and type 2 diabetes,[8] suggesting a significant link between low plasma adiponectin levels and insulin resistance. Low plasma adiponectin is also associated with hepatic steatosis and hepatic insulin resistance in NAFLD.[9] Adiponectin circulates in the plasma in 3 forms: low-molecular-weight trimer, a hexamer (trimer-dimer) of medium molecular weight, and a larger multimeric high-molecular-weight (HMW) form.[10] Of these, HMW adiponectin is believed to be the active form of this adipokine.[11] In a previous study, we demonstrated that the serum HMW adiponectin level is profoundly reduced in patients with type 2 diabetes complicated by coronary artery diseases.[12] A meta-analysis and our previous studies confirmed that SGLT2 inhibitors increase modestly but significantly serum total or HMW adiponectin in patients with type 2 diabetes.[7 13 14] It is possible that the increase in serum adiponectin after treatment with SGLT2 inhibitors improves insulin sensitivity and thereby lowers hyperglycemia in patients with type 2 diabetes. We hypothesize that SGLT2 inhibitors may be more effective in patients with type 2 diabetes and NAFLD whose serum HMW adiponectin is low. Based on the hypothesis that serum HMW adiponectin level is a predictor of response to dapagliflozin in patients with type 2 diabetes complicated by NAFLD, we investigated whether the baseline serum HMW adiponectin level was associated with the response to treatment with dapagliflozin for 24 weeks in patients with type 2 diabetes and NAFLD.

SUBJECTS AND METHODS

The design and primary study results of our original trial have been reported.[7] In brief, this was a prospective, randomized, and open-label study of 63 patients with type 2 diabetes and NAFLD who were referred to the diabetes outpatient clinic of Dokkyo Medical University. Eligible patients had type 2 diabetes with NAFLD, were at least 20 years old, and had an HbA1c level of 6.0%–12.0%, and were on stable dosages of 1–3 oral antidiabetic agents with or without insulin for at least 3 months. Patients with renal insufficiency (estimated GFR <45 mL/min/1.73 m^2), or any liver diseases other than NAFLD were excluded. NAFLD was diagnosed on the basis of liver dysfunction (persistent elevation of alanine transaminase ≥ institutional upper limit of normal), fatty liver on ultrasonography, and low daily alcohol intake (less than 30 g in men or less than 20 g in women). We also excluded patients treated with glucagon-like peptide-1 receptor agonists because it may have an impact on hepatic steatosis.

Patients were randomly assigned to receive either dapagliflozin or standard treatment without SGLT2 inhibitors (the control group). Each patient was followed for 24 weeks with monthly review. The dose of dapagliflozin was fixed at 5 mg/d, the standard dose for type 2 diabetes in Japan. The control group received standard treatment for type 2 diabetes, and if the HbA1c target (less than 7.0%) is not achieved after approximately 3 months, uptitration treatment was done with antidiabetic drugs excluding SGLT2 inhibitors. As additional antidiabetic drugs in the control group, 3 patients were newly started with dipeptidyl peptidase-4 inhibitors, 2 patients with alpha glucosidase inhibitors, 1 patient with glinides, and 1 patient with basal insulin during the study period.

All patients received standard-of-care treatment for type 2 diabetes throughout the study, and investigators treated the patients according to local guidelines to achieve effective glycemic control.[7]

The visceral fat area (VFA) was defined as the area under the curve for the umbilical section on dual bioelectrical impedance analysis (Dual Scan, Omron Healthcare, Kyoto, Japan). This instrument calculates the cross-sectional area

Table 2 Changes in clinical parameters in patients with type 2 diabetes and NAFLD treated with dapagliflozin or control (standard treatment)

	Dapagliflozin			Control		
	Baseline	Week 24	P value	Baseline	Week 24	P value
n	33	33	–	24	24	–
Body weight (kg)	73.6 (61.9, 80.8)	70.7 (60.0, 79.2)	0.0004	74.9 (65.6, 81.6)	76.9 (65.7, 82.3)	0.9429
BMI (kg/m^2)	27.6±4.7	26.9±5.0	0.0006	28.7±3.5	28.6±3.6	0.4930
Body fat (kg)	23.5±9.1	22.9±8.7	0.1186	26.8±8.9	26.4±8.8	0.3468
VFA (cm^2)	108.7±42.9	101.4±39.2	0.0068	125.7±32.2	120.0±40.1	0.1795
Body water (kg)	37.2±5.0	36.2±9.5	0.0057	36.8±6.7	36.5±6.4	0.9762
ECW/TBW	0.387±0.008	0.387±0.008	0.7729	0.388±0.009	0.388±0.009	0.9606
FPG (mg/dL)	137.8±54.0	122.5±35.2	0.1057	136.5±41.2	144.3±45.5	0.2218
HbA1c (%)	8.37±1.48	7.36±1.22	<0.0001	7.70±1.24	7.22±1.11	0.1414
HOMA-IR	3.58 (2.41, 6.10)	2.66 (1.49, 4.85)	0.0076	3.92 (1.96, 5.47)	3.56 (2.17, 5.78)	0.9240
Fasting C peptide (ng/mL)	1.8 (1.03, 2.63)	1.5 (1.03, 2.3)	0.2620	1.95 (1.23, 2.88)	2.3 (1.38, 2.95)	0.5883
LDL cholesterol (mg/dL)	108.1±29.5	109.4±32.1	0.6398	102.4±23.6	109.3±23.5	0.2842
Triglyceride (mg/dL)	132.6±54.7	117.5±48.2	0.0842	144.8±92.5	141.7±84.6	0.3512
HDL cholesterol (mg/dL)	49.7±12.8	55.0±14.7	0.0011	46.1±12.2	47.8±11.0	0.1126
AST (U/L)	28.0 (20.5, 49.8)	27.5 (17.3, 31.8)	0.0018	29.8±12.8	27.4±9.6	0.3353
ALT (U/L)	38.0 (21.5, 61.0)	26.5 (16.3, 42.5)	<0.0001	33.0 (24.5, 46.5)	32.0 (25.0, 49.3)	0.4493
GGT (U/L)	47.0 (28.0, 88.3)	27.0 (20.5, 61.5)	0.0003	37.5 (20.0, 62.3)	32.0 (22.3, 50.0)	0.4584
Uric acid (mg/dL)	4.80±1.24	4.46±1.20	0.0236	5.16±1.33	5.12±1.23	0.7527
Hematocrit (%)	44.3±3.9	45.6±4.3	0.0002	44.2±3.7	44.8±3.2	0.3864
eGFR (mL/min/1.73 m^2)	79.4±15.8	82.3±17.1	0.1435	76.9±19.0	80.8±24.0	0.1586
HMW adiponectin (μg/mL)	1.08 (0.56, 4.11)	1.62 (0.91, 4.67)	0.0002	1.31 (0.44, 2.53)	1.79 (0.39, 2.55)	0.3894
Type 4 collagen 7S (ng/mL)	4.00 (3.53, 4.90)	4.00 (3.32, 4.78)	0.4707	3.71±0.91	4.15±1.06	0.0295
CAP (dB/m)	314.6±61.0	290.3±72.7	0.0424	306.0±34.3	311.3±37.3	0.6253
Metformin/DPP-4i/SU/Ins	31/16/12/10			21/14/11/4		

Data are mean±SD or the median and IQRs.
ALT, alanine transaminase; AST, aspartate aminotransferase; BMI, body mass index; CAP, controlled attenuation parameter; DPP, dipeptidyl peptidase; ECW, extracellular water; eGFR, estimated glomerular filtration rate; FPG, fasting plasma glucose; GGT, γ-glutamyltranspeptidase; HDL, high-density lipoprotein; HMW, high-molecular-weight; HOMA-IR, homeostasis model assessment of insulin resistance; Ins, insulin; LDL, low-density lipoprotein; NAFLD, non-alcoholic fatty liver disease; SU, sulfonylurea; TBW, total body water; VFA, visceral fat area.

of intra-abdominal fat (visceral adipose tissue and subcutaneous adipose tissue) at the umbilicus based on measurement of electrical potentials after application of small electrical currents to 2 different body spaces.[15] Body composition was measured with a bioimpedance analysis device (InBody 720). Extracellular water (ECW) and total body water (TBW) were calculated using the body volume model. The severity of hepatic steatosis was evaluated by the controlled attenuation parameter (CAP) using transient elastography (FibroScan; Echosens, Paris, France), which is expressed in dB/m.

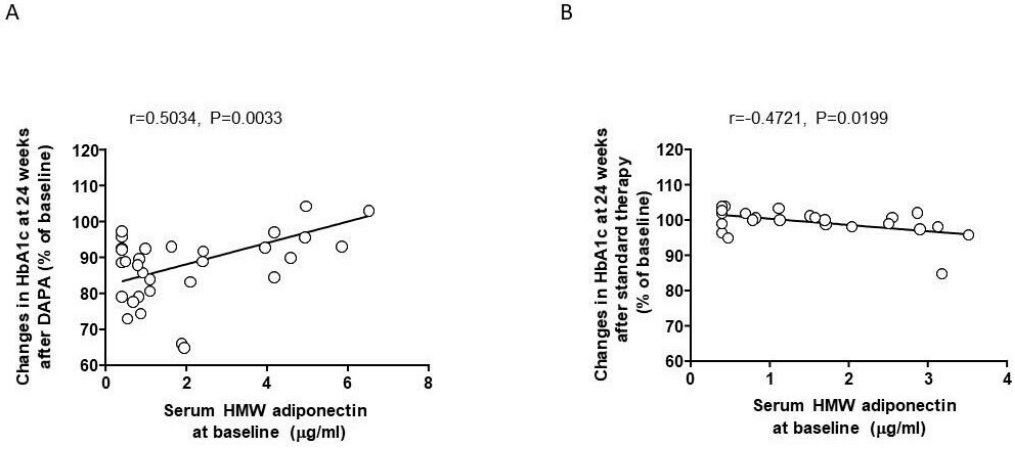

Figure 1 Correlation between baseline serum high-molecular-weight (HMW) adiponectin and changes in HbA1c from baseline to 24 wk of treatment with dapagliflozin (A) or with standard therapy (B) in patients with type 2 diabetes and non-alcoholic fatty liver disease (NAFLD). DAPA, dapagliflozin.

Table 3 Univariate and multivariate analyses of relationships between changes in HbA1c from baseline to 24 wk after treatment with dapagliflozin and baseline clinical variables in 33 patients with type 2 diabetes and NAFLD

	Univariate analysis		Multivariate analysis	
Variable value	r	P value	β	P value
Age (y)	0.3367	0.0544	NE	
Body weight (kg)	−0.3987	0.0238	NE	
BMI	−0.3764	0.0337	NE	
Total fat mass (kg)	−0.4052	0.0214	NE	
VFA (cm^2)	−0.3613	0.0422	−0.128	0.510
Body water (kg)	−0.2155	0.2362	NE	
ECW/TBW	0.2127	0.2426	NE	
FPG (mg/dL)	−0.2336	0.1982	NE	
HbA1c (%)	−0.4659	0.0063	−0.559	0.002
HOMA-IR	−0.1635	0.3713	NE	
Fasting serum C peptide (ng/mL)	−0.2539	0.1609	NE	
LDL cholesterol (mg/dL)	−0.3463	0.0522	NE	
Triglyceride (mg/dL)	0.0640	0.7280	NE	
HDL cholesterol (mg/dL)	0.1867	0.3062	NE	
eGFR (mL/min/1.73 m^2)	−0.2755	0.1270	NE	
Uric acid (mg/dL)	0.1277	0.4935	NE	
AST (U/L)	−0.0852	0.6428	NE	
ALT (U/L)	−0.2726	0.1312	NE	
GGT (U/L)	−0.3261	0.0686	NE	
hs-CRP	−0.4010	0.0229	NE	
HMW adiponectin (μg/mL)	0.5034	0.0033	0.471	0.010
Type 4 collagen 7S (ng/mL)	−0.4335	0.0132	NE	
CAP	−0.2126	0.2594	NE	
R^2			0.485	

β indicates partial coefficient.
ALT, alanine transaminase; AST, aspartate aminotransferase; BMI, body mass index; CAP, controlled attenuation parameter; ECW, extracellular water; eGFR, estimated glomerular filtration rate; FPG, fasting plasma glucose; GGT, γ-glutamyltranspeptidase; HDL, high-density lipoprotein; HMW, high-molecular-weight; HOMA-IR, homeostasis model assessment of insulin resistance; hs-CRP, high-sensitivity C-reactive protein; LDL, low-density lipoprotein; NAFLD, non-alcoholic fatty liver disease; NE, does not enter the final model; TBW, total body water; VFA, visceral fat area.

Serum HMW adiponectin was measured with our sandwich ELISA employing a monoclonal antibody targeting human HMW adiponectin, as described previously.[12] Serum type 4 collagen 7S level was determined by using radioimmunoassay kits (Human type 4 collagen 7S kit, Sceti Medical Labo, Tokyo, Japan). All included subjects gave informed consent for participation in this study. This study was registered with University Hospital Medical Information Network Clinical Trials Registry.

Statistical analysis

Results are expressed as the mean±SD or the median and IQR. Differences between groups were analyzed by Student's paired t-test or the unpaired t-test, while differences in non-parametric data were assessed by Wilcoxon matched-pairs test or the Mann-Whitney U test. Correlations were determined by linear regression analysis or Spearman's rank method. To identify independent determinants of changes (reductions) in HbA1c from baseline to 24 weeks after treatment with dapagliflozin, we performed multivariate logistic regression analysis that included 3 significant variables evaluated by linear regression analysis, because sample size was calculated as the number of variables multiplied by 10 in multiple logistics regression analysis. Statistical analyses were carried out using SPSS V.8.0 J software (SPSS), and $p<0.05$ was accepted as indicating statistical significance.

RESULTS

A total of 63 patients were screened and underwent randomization to receive dapagliflozin (n=35) or standard treatment (n=28). In the dapagliflozin group, 33/35 patients completed the trial, while 24/28 patients completed it in the standard treatment group.

In a total of 57 patients with type 2 diabetes and NAFLD, the baseline serum level of HMW adiponectin showed a positive correlation with ECW/TBW and high-density lipoprotein (HDL) cholesterol (table 1), whereas it was negatively correlated with body weight, TBW, VFA, serum C peptide, triglyceride (TG), liver enzymes, hematocrit, and CAP. According to the multivariate analysis, only ECW/TBW (β=0.349, p=0.027) and HDL cholesterol (β=0.365, p=0.024) were independent factors for serum HMW adiponectin level at baseline (table 1).

At baseline, the 2 groups were well balanced with respect to demographic characteristics and laboratory data (table 2). HbA1c decreased significantly from 8.37%±1.48% at baseline to 7.36%±1.22% after 24 weeks of dapagliflozin treatment (table 2). HbA1c also decreased in the standard treatment (control) group, but the change was not significant. Serum HMW adiponectin showed a significant increase at 24 weeks in the dapagliflozin group, but not in the control group (table 2). VFA decreased significantly by the end of treatment in the dapagliflozin group, while there were no changes in these parameters in the control group (table 2). CAP was significantly decreased after 24 weeks in the dapagliflozin group, while there was no change in CAP in the control group (table 2).

In the dapagliflozin group, the baseline serum HMW adiponectin was positively correlated with reductions in HbA1c from baseline to 24 weeks after treatment (figure 1A; r=0.5034, p=0.0033), while in the control group, the baseline serum HMW adiponectin was negatively correlated with reductions in HbA1c after treatment (figure 1B; r=−0.4721, p=0.0199) (table 3). In the dapagliflozin group, the baseline body weight, BMI, VFA, HbA1c, high-sensitivity C-reactive protein, and type 4 collagen 7S were negatively correlated with reductions in HbA1c after treatment (online supplemental table 1). On the other hand, the baseline HMW adiponectin showed no significant correlation with changes in CAP (r=0.128, p=0.510).

According to multivariate logistic regression analysis, in a model that explained 69% (R^2=0.485) of the variation of the changes (reductions) in HbA1c from baseline to 24 weeks after treatment with dapagliflozin, the baseline HbA1c (β=−0.559, p=0.002) and serum HMW

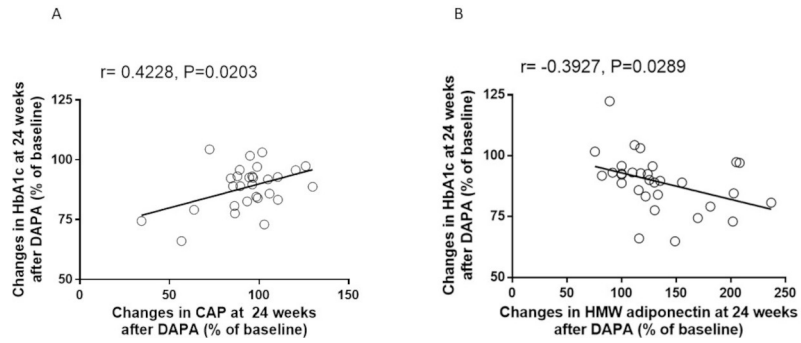

Figure 2 Correlation between changes in HbA1c and those in controlled attenuation parameter (CAP) (A) or those in high-molecular-weight (HMW) adiponectin (B) at 24 wk of treatment with dapagliflozin (DAPA) in patients with type 2 diabetes and non-alcoholic fatty liver disease (NAFLD).

adiponectin (β=0.471, p=0.010) were independent determinants of the reduction of HbA1c at 24 weeks (table 3). In the control group, the baseline HbA1c was also negatively correlated with the reduction in HbA1c after treatment with dapagliflozin.

In the dapagliflozin group, the change in HbA1c was positively correlated with the changes in CAP (figure 2A) and serum type 4 collagen 7S (online supplemental table 1), while it was negatively correlated with the change in TBW (Online supplemental table 1) and HMW adiponectin (figure 2B). In the control group, there was no significant correlation between changes in HbA1c and those in HMW adiponectin at 24 weeks after treatment (r=−0.060, p=0.8087; data not shown).

DISCUSSION

In the present study, we showed for the first time that the baseline serum level of HMW adiponectin was positively correlated with changes in HbA1c from baseline to 24 weeks with dapagliflozin (5 mg/d) treatment in patients with type 2 diabetes who had NAFLD. In contrast, there was a negative correlation in the standard therapy group between baseline HMW adiponectin and changes in HbA1c after treatment. Furthermore, in the multivariate regression analysis, the baseline serum HMW adiponectin was an independent determinant of the reductions of HbA1c at 24 weeks after treatment with dapagliflozin. Thus, a low baseline serum HMW adiponectin level may be associated with a better response to dapagliflozin in patients with type 2 diabetes and NAFLD.

The mechanisms underlying the relationship between a lower baseline serum HMW adiponectin level and a better response of HbA1c to treatment with dapagliflozin remain to be determined. At baseline serum HMW adiponectin showed a negative correlation with VFA and CAP in a total of 57 patients, indicating that low serum adiponectin is associated with visceral fat accumulation and/or hepatic steatosis in patients with type 2 diabetes, both of which induce insulin resistance in the target organs of insulin action. Low adiponectin in NAFLD is also correlated negatively with hepatic insulin sensitivity and positively with the amount of hepatic fat content, resulting in an impaired suppression of endogenous glucose production by insulin.[16] It is known that SGLT2 inhibitors promote weight reduction due to fat loss caused by a negative effect on the energy balance, and also volume loss caused by its diuretic effect, or due to a combination of both.[1 2] A recent study found that body weight reduction with SGLT2 inhibitor treatment was caused mainly by a decrease in adipose tissue mass after 6 months (long term), while only a transient decrease of ECW was induced from 3 days to 1 month (short term).[17] Several studies, including ours, have demonstrated that SGLT2 inhibitors reduced hepatic steatosis in patients with type 2 diabetes complicated by NAFLD.[7 18 19] Using a mouse model of NAFLD-NASH with diabetes, we showed that SGLT2 inhibitors reduced ectopic accumulation of lipids (TG) in the liver by promoting fat oxidation and inhibiting de novo lipogenesis,[20] resulting in an improvement of NAFLD. Considering all the above, one possible explanation is that because dapagliflozin promotes fat loss in both adipose tissue (especially visceral fat) and the liver, and also improves insulin sensitivity in patients with type 2 diabetes and NAFLD, dapagliflozin is more effective in subjects with higher insulin resistance whose serum HMW adiponectin was low. Thus, SGLT2 inhibitors reduce plasma glucose in the long term by improving insulin resistance via fat loss in both the liver and adipose tissues.

In the present study, the change in HbA1c showed a significant negative correlation with the changes in HMW adiponectin after treatment with dapagliflozin. To our knowledge, this is the first study that shows that an increase in serum HMW adiponectin by dapagliflozin treatment may contribute to an improvement of glycemic control in patients with type 2 diabetes and NAFLD. Serum HMW adiponectin showed a significant increase at 24 weeks in the dapagliflozin group, but not in the standard treatment (control) group. These results suggest that a more considerable increase in serum HMW adiponectin by dapagliflozin may be associated with a more substantial reduction in HbA1c after 24 weeks of dapagliflozin treatment in patients with type 2 diabetes and NAFLD.

The present study demonstrated that dapagliflozin significantly decreased CAP (a marker of hepatic steatosis) as evaluated by FibroScan. We also found a significant positive correlation between changes in HbA1c and those in CAP after treatment with dapagliflozin, suggesting that dapagliflozin improves glycemic control by ameliorating insulin sensitivity via a reduction of fat in the liver. In a previous study, we demonstrated that empagliflozin markedly reduced TG content in the liver by inhibiting fatty acid

production (de novo lipogenesis) in addition to an increase of fatty acid oxidation in a mice model of NASH.[20] Dapagliflozin may decrease HbA1c level by fat loss in both visceral adipose tissue and the liver in people with type 2 diabetes and NAFLD, an underlying mechanism supported by the present study showing a significant decrease in VFA and CAP after treatment with dapagliflozin.

The multivariate analysis in this study found baseline HbA1c to be an independent determinant of the reduction of HbA1c after 24-week treatment, confirming that high baseline HbA1c is associated with a greater reduction in HbA1c after treatment with dapagliflozin in this patient group, in agreement with several previous reports.[4–6] Ferrannini *et al*[3] demonstrated that SGLT2 inhibitor ipragliflozin increases glucosuria in direct proportion to GFR and fasting plasma glucose in patients with type 2 diabetes, suggesting that the higher the degree of hyperglycemia the better SGLT2 inhibitors improve HbA1c. However, because high baseline HbA1c (before treatment) is associated with a greater change in HbA1c with most antidiabetic drug treatment, the relationship between baseline HbA1c and the glucose-lowering effect of antidiabetic drugs may not be specific to SGLT2 inhibitors.

In conclusion, a lower serum HMW adiponectin level was associated with a better response to dapagliflozin in patients with type 2 diabetes and NAFLD.

Contributors MS, TN, and KK contributed to the study design, data collection, and drafting of the manuscript. TI, TJ, and TT contributed to the discussion and reviewed the manuscript. IU reviewed/edited the manuscript. YA researched the data, and wrote and reviewed/edited the manuscript.

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Competing interests None declared.

Patient consent for publication Not required.

Ethics approval The present study was approved by the Institutional Review Board of Dokkyo Medical University Hospital (protocol number 27158).

Provenance and peer review Not commissioned; externally peer reviewed.

Data availability statement Data are available in a public, open-access repository.

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