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Comparison of tofogliflozin 20 mg and ipragliflozin 50 mg used together with insulin glargine 300 U/mL using continuous glucose monitoring (CGM): A randomized crossover study

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Abstract. To investigate whether sodium glucose co-transporter 2 inhibitors (SGLT2i), tofogliflozin or ipragliflozin, achieve optimal glycemic variability, when used together with insulin glargine 300 U/mL (Glargine 300). Thirty patients with type 2 diabetes were randomly allocated to 2 groups. For the first group: After admission, tofogliflozin 20 mg was administered; Fasting plasma glucose (FPG) levels were titrated using an algorithm and stabilized at 80 mg/dL level with Glargine 300 for 5 days; Next, glucose levels were continuously monitored for 2 days using continuous glucose monitoring (CGM); Tofogliflozin was then washed out over 5 days; Subsequently, ipragliflozin 50 mg was administered; FPG levels were titrated using the same algorithm and stabilized at 80 mg/dL level with Glargine 300 for 5 days; Next, glucose levels were continuously monitored for 2 days using CGM. For the second group, ipragliflozin was administered prior to tofogliflozin, and the same regimen was maintained. Glargine 300 and SGLT2i were administered at 8:00 AM. Data collected on the second day of measurement (mean amplitude of glycemic excursion [MAGE], average daily risk range [ADRR]; on all days of measurement) were analyzed. Area over the glucose curve (<70 mg/dL; 0:00 to 6:00, 24-h), M value, standard deviation, MAGE, ADRR, and mean glucose levels (24-h, 8:00 to 24:00) were significantly lower in patients on tofogliflozin than in those on ipragliflozin. Tofogliflozin, which reduces glycemic variability by preventing nocturnal hypoglycemia and decreasing postprandial glucose levels, is an ideal SGLT2i when used together with Glargine 300 during basal insulin therapy.

Key words: Tofogliflozin, Insulin glargine 300 U/mL, Continuous glucose monitoring

BASAL INSULIN THERAPY, in which the patient's preferences and view-points are considered, is effective and safe, and has been commonly used as a part of insulin therapy. Basal insulin therapy can support basal and bolus insulin secretion by combining once-daily dosage of basal insulin and oral hypoglycemic agents (OHA). In the ideal basal insulin therapy, basal insulin is used to decrease fasting plasma glucose (FPG) levels, reducing the occurrence of hypoglycemia, while an oral hypoglycemic agent is used to decrease postprandial glucose levels, reducing the occurrence of hypoglycemia.

A relationship between nocturnal hypoglycemia and sudden death has been suggested [1]. As per the Somogyi phenomenon, nocturnal hypoglycemia causes an increase in the difference between pre- and post-breakfast glucose levels [2], and this leads to an increase in daytime glucose levels. Thus, hypoglycemia and increased glycemic variability occur at the same time during the Somogyi phenomenon. Large clinical studies have shown that hypoglycemia and glycemic variability are associated with mortality in patients with diabetes mellitus [3-5]. We wanted to predict the Somogyi phenomenon, in which hypoglycemia and increased glycemic variability occur concomitantly, and we have previously reported that major increases between pre- and post-breakfast glucose levels may predict nocturnal hypoglycemia in type 2 diabetes [6].

When the Somogyi phenomenon is suspected, reducing both nocturnal hypoglycemia and increases in daytime glucose levels through increases between pre- and post-breakfast glucose levels, establishing an appropriate control of pre-breakfast glucose levels, is necessary [1, 4, 7]. We believe that long-acting insulin to reduce nocturnal hypoglycemia, and OHA to reduce

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daytime (postprandial) glucose levels efficiently is useful in basal insulin therapy.

Sodium glucose co-transporter 2 inhibitors (SGLT2i) are useful OHAs that decrease glucose levels in a glucose-level dependent manner with a single agent [8] and that leads to many second order effect, such as decrease in body weight and blood pressure [9]. It has been reported that tofogliflozin taken in the morning decreases postprandial glucose levels of about 70 mg/dL and FPG of about 35 mg/dL [10]. In contrast, It has also been reported that ipragliflozin decreases postprandial glucose levels of about 60 mg/dL and FPG of about 45 mg/dL [11]. From those reports, we can estimate that the effect of tofogliflozin is stronger during the daytime and weaker during the nighttime than that of ipragliflozin even though there are no differences between the overall effects of tofogliflozin or ipragliflozin. Thus, an optimal SGLT2i used in basal insulin therapy, tofogliflozin taken in the morning may decrease postprandial glucose levels more than ipragliflozin. In addition, SGLT2i when used together with insulin, increases the risk of hypoglycemia. Therefore, when SGLT2i is used together with insulin, if the effect of the SGLT2i remains at nighttime, the risk of hypoglycemia is thought to increase, because of the combined effect of SGLT2i and insulin. It has been reported that tofogliflozin has the shortest half-life among all other SGLT2i [12] and that a rate of urinary tofogliflozin excretion is more than 80 % at the time of 12 h after administration [12]. It has also been reported that a rate of urinary tofogliflozin excretion is more than 96 % at the time of 24 h after administration and only about 3 % during 24 h ~ 48 h after administration [12]. Thus, morning administeration of tofogliflozin may reduce the risk of hypoglycemia because the SGLT2i's effects almost disappear by nighttime and there are almost no overcarrying effect since 24 h after administration.

Regarding reduction of nocturnal hypoglycemia, it was observed that nocturnal and overall occurrence of hypoglycemic events were significantly reduced in patients on insulin glargine 300 U/mL (Glargine 300), as opposed to insulin glargine 100 U/mL [13]. Therefore, Glargine 300 is thought to be a useful long-acting insulin for reducing nocturnal hypoglycemia.

Thus, combination therapy using Glargine 300 (considered to help in decreasing pre-breakfast glucose levels, reducing the occurrence of nocturnal hypoglycemia) and tofogliflozin (considered to help in reducing the occurrence of nocturnal hypoglycemia when used together with insulin and in decreasing postprandial glucose levels) administered in the morning, may be an ideal basal insulin therapy using SGLT2i. To investigate whether tofogliflozin decreases the occurrence of nocturnal hypoglycemia and postprandial glucose levels when used together with insulin, we compared the effects of tofogliflozin 20 mg + Glargine 300, to that of ipragliflozin 50 mg + Glargine 300, using continuous glucose monitoring (CGM) in a randomized crossover study.

Materials and Methods

Study design and patient selection

Nocturnal urinary frequency of the ipragliflozin group is 1.7 ± 0.7 times, as reported by Yoshida in a clinical trial investigating urinary frequency of patients administered tofogliflozin [14]. Thirty individuals were required in each control group and treatment group, in order to be able to detect a 30% alteration in the mean nocturnal urinary frequency, with an α value of 0.05 and with a statistical power of 80%. We therefore determined that the total number of participants required was 30.

Thirty patients with type 2 diabetes, treated with Glargine 300 during basal insulin therapy for 3 months or longer, were randomly allocated into 2 groups. For the first group (T/I group): After admission, tofogliflozin 20 mg was administered once daily; FPG levels were titrated using an algorithm made refering to a clinical trial [FPG levels on the day of dosage adjustment, dosage adjustment of Glargine 300 (U/day): FPG levels ≥ 180 mg/dL, +4; 140 mg/dL ≤ FPG levels < 180 mg/dL, +3; 110 mg/dL ≤ FPG levels < 140 mg/dL, +2; 90 mg/dL ≤ FPG levels < 110 mg/dL, +1; 80 mg/dL ≤ FPG levels < 90 mg/dL, no change; FPG levels < 80 mg/dL, -2; adjustment on every second day] [13] and stabilized at 80 mg/dL level with Glargine 300 for 5 days; Next, glucose levels were continuously monitored for 2 days using the continuous glucose monitoring (CGM) device; Tofogliflozin was then washed out over the next 5 days; Subsequently, ipragliflozin 50 mg was administered once daily; FPG levels were titrated using the same algorithm and stabilized at 80 mg/dL level with Glargine 300 for 5 days; Next, glucose levels were continuously monitored for 2 days using the CGM device. For the second group (I/T group), ipragliflozin was administered prior to

tofogliflozin, and the same regimen was maintained (Fig. 1). Patients were allocated to groups using a random number table, and the study design was continuous and prospective. Glargine 300 and SGLT2i were administered at 8:00 AM. The Glargine 300 dose was adjusted to be constant no later than start of continuous glucose monitoring. The Glargine 300 dose was constant during the CGM measurement period. FPG levels were titrated using the same algorithm and stabilized at 80 mg/dL level with Glargine 300 during washing out. Data collected on the second day of measurement (mean amplitude of glycemic excursion (MAGE) [15], mean of daily differences (MODD) [16], average daily risk range (ADRR) [17]: all days of measurement) were analyzed. Regarding other diabetic treatments, sulfonylurea agents, α- glucosidase inhibitors, rapid-acting insulin secretagogues, and glucagon-like peptide-1 receptor agonists were discontinued after enrollment. All other treatments were continued during the research period. The total caloric intake for subjects was defined by the attending physicians as 1,440 kcal, 1,600 kcal, or 1,840 kcal per day, to accommodate differences in physique among the subjects [18]. Test meals were given to each patient, based on recommendations by the Japan Diabetes Society [component ratio of calories (carbohydrates 60%, proteins 18%, and lipids 22%), (breakfast 30% of total calories, lunch 35% of total calories, and supper 35% of total calories)], irrespective of differences in physique during the CGM measurement period. Physical activity during the reserch period was 1.5 metabolic equivalents, based on the analysis of baseline data. Patients whose estimated glomerular filtration rate was less than 45 mL/min/1.73m^2 or were judged to be unsuitable for participation for other medical reasons, were excluded from this study.

Patients who provided informed concent were included in this study and we performed this study based on Helsinki Declaration. The study protocol was approved by the Ethical Committee of General Inuyamachuo Hospital (authorization no. III, 2016), and was registered in a clinical trial database with the University Medical Information Network (no. UMIN000023972).

Outcomes and statistical analysis

Nighttime was defined as 0:00-6:00 and hypoglycemia was defined as glucose level less than 70 mg/dL [13].

The primary CGM endpoint included area over the glucose curve (AOC) (<70 mg/dL) (0:00-6:00, 24 h), M-value (0:00-6:00 (target glucose level = 90)) [19], MAGE, ADRR, mean glucose level (8:00-24:00), standard deviation (SD) (0:00-6:00, 8:00-24:00) [20].

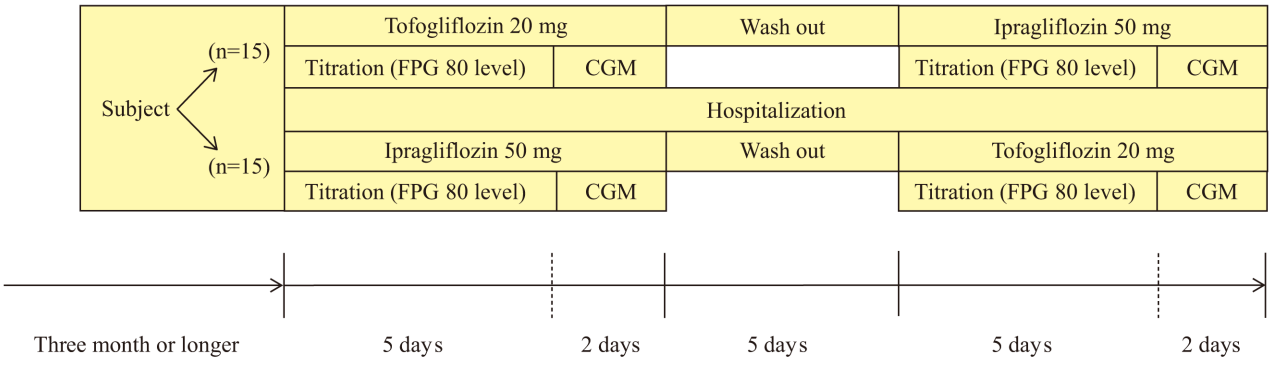

Fig. 1 Study protocol

Thirty patients with type 2 diabetes were randomly allocated to 2 groups. For the first group: After admission, tofogliflozin 20 mg was administered; Fasting plasma glucose (FPG) levels were titrated using an algorithm and stabilized at 80 mg/dL level (80 level) with Glargine 300 for 5 days; Next, glucose levels were continuously monitored for 2 days using the continuous glucose monitoring (CGM) device; Tofogliflozin was then washed out over the next 5 days; Subsequently, ipragliflozin 50 mg was administered; FPG levels were titrated using the same algorithm and stabilized at 80 level with Glargine 300 for 5 days; Next, glucose levels were continuously monitored for 2 days using the CGM device. For the second group, ipragliflozin was administered prior to tofogliflozin, and the same regimen was maintained. The Glargine 300 dose was adjusted to be constant no later than start of continuous glucose monitoring. The Glargine 300 dose was constant during the CGM measurement period.

The secondary CGM endpoint included AOC (<70 mg/dL) (8:00-24:00), M-value (24 h (target glucose level = 100), 8:00-24:00 (target glucose level = 120)) [19], MODD, mean glucose level (24 h, 0:00-6:00), SD (24 h) [20], area under the glucose curve (AUC) (≥140 mg/dL) within 3 hours of each meal [21], and AUC (≥0 mg/dL) in 24 hours [21]. The secondary urinary endpoint included urinary glucose level (0:00-6:00, 6:00-24:00, 24 h), urinary frequency (0:00-6:00, 6:00-24:00, 24 h) and urinary volume (0:00-6:00, 6:00-24:00, 24 h).

Data are shown as median (interquartile range). Statistical analysis was performed with Wilcoxon signed-rank test. A p value of less than 0.05 was considered significant. The data were analyzed using the BellCurve for Excel software program (Social Survey Research Information Co., Ltd.).

Results

Clinical characteristics of the patient

Table 1 shows the patients' clinical characteristics. The study included 20 men and 10 women. The baseline characteristics were as follows: age, 73.0 (66.0-79.0) years; duration of diabetes, 10.0 (5.0-19.5) years; Body Mass Index (BMI), 23.1 (19.7-24.9) kg/m^2; HbA1c, 8.3 (7.4-8.7) %; C-peptide immunoreactivity (CPR), 1.2 (0.6-1.7) ng/mL; FPG level, 123.0 (94.5-147.0) mg/dL; C-peptide index, 0.8 (0.5-1.2); urine-CPR, 25.6 (14.2-45.0) μg/day; and estimated glomerular filtration rate (eGFR), 54.2 (46.6-72.6). All the characteristics were not significantly different between T/I group and I/T group (Table 1).

Outcomes

Fig. 2 shows the glycemic variability over 24 hours of CGM in all patients.

The results of primary endpoint are shown as follows: AOC (<70 mg/dL) (0:00-6:00, 24 h) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.001, p=0.0002, respectively); M value (0:00-6:00) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p<0.0001); MAGE was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.0003); ADRR was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.001); Mean glucose levels (8:00-24:00) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.0001); SD (0:00-6:00, 8:00-24:00) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.0007, p=0.0001, respectively).

Table 1 Baseline characteristics

Characteristic	Total	T/I group	I/T group	p (T/I *versus* I/T)
N (Male / Female)	30 (20 / 10)	15 (9 / 6)	15 (11 / 4)	p_2=0.44
Age, years	73.0 (66.0-79.0)	74.0 (66.5-79.5)	71.0 (59.5-77.5)	p_1=0.41
Duration of diabetes, years	10.0 (5.0-19.5)	10.0 (7.0-15.5)	7.0 (4.5-22.0)	p_1=0.53
BMI, kg/m^2	23.1 (19.7-24.9)	23.9 (20.8-25.5)	21.4 (19.3-24.7)	p_1=0.47
HbA1c (NGSP), %	8.3 (7.4-8.7)	8.4 (7.3-8.8)	7.8 (7.5-8.6)	p_1=0.88
GA, %	22.7 (17.2-27.1)	22.2 (18.2-25.3)	24.1 (17.2-28.5)	p_1=0.66
CPR, ng/mL	1.2 (0.6-1.7)	1.2 (0.7-1.7)	1.2 (0.6-1.7)	p_1=0.88
FPG, mg/dL	123.0 (94.5-147.0)	122.0 (94.0-160.5)	124.0 (105.5-136.5)	p_1=0.77
CPI	0.8 (0.5-1.2)	0.8 (0.7-1.3)	0.9 (0.4-1.1)	p_1=0.98
U-CPR, μg/day	25.6 (14.2-45.0)	21.8 (13.6-42.5)	26.2 (14.2-44.4)	p_1=0.71
eGFR, mL/min/1.73m^2	54.2 (46.6-72.6)	55.6 (45.8-67.0)	54.2 (46.7-84.5)	p_1=0.71
Biguanide agent, *n* (%)	18 (60.0)	8 (53.3)	10 (60.0)	p_2=0.46
Thiazolidine, *n* (%)	3 (10.0)	3 (10.0)	0 (0)	p_2=0.07
DPP4 inhibitor, *n* (%)	23 (76.7)	12 (80.0)	11 (73.3)	p_2=0.67
ACE inhibitor, *n* (%)	1 (6.7)	1 (6.7)	0 (0)	p_2=0.31
ARB, *n* (%)	9 (30.0)	4 (26.7)	5 (33.3)	p_2=0.69
Statin, *n* (%)	7 (23.3)	2 (13.3)	5 (33.3)	p_2=0.2

Data are shown as median (interquartile range). p_1: Mann-Whitney's U test, p_2 : Chi-square test. BMI, body mass index; HbA1c, glycosylated hemoglobin; GA, glycoalbumin; CPR, C-peptide immunoreactivity; FPG, fasting plasma glucose; CPI, C-peptide index; U-CPR, urine-CPR; eGFR, estimated glomerular filtration rate; DPP, dipeptidyl-peptidase; ACE inhibitor, angiotensin-converting enzyme inhibitor; ARB, angiotensin II receptor blocker; T/I, Tofogliflozin (first) / Ipragliflozin (second); I/T, Ipragliflozin (first) / Tofogliflozin (second).

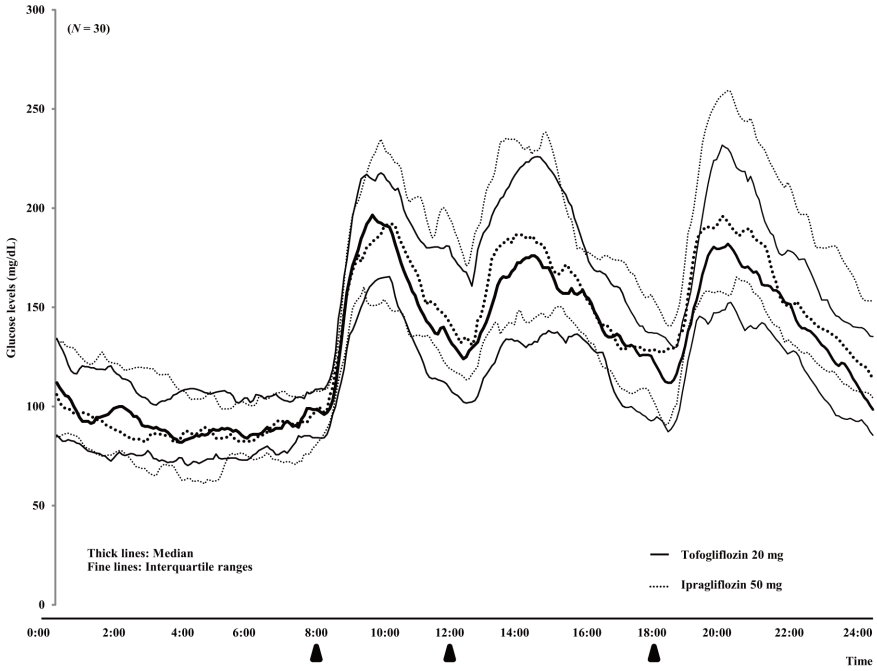

Fig. 2 The graph indicates glycemic variability over 24 h on CGM in patients during treatment with tofogliflozin 20 mg + Glargine 300 or ipragliflozin 50 mg + Glargine 300. Glucose levels were calculated from the value of CGM on the second measurement day. Data are shown as median (thick lines) and interquartile ranges (fine lines).

The results of secondary endpoint are shown as follows: M value (24 h, 8:00-24:00) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.008, p=0.0003, respectively); Mean glucose levels (24 h) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.009); SD (24 h) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.0002); AUC (≥140 mg/dL) within 3 hours after every meal was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.02, p=0.003, p=0.0003, respectively); AUC (≥0 mg/dL) in 24 hours was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.007); Urinary glucose level (0:00-6:00, 24 h) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.0002, p=0.03, respectively); Urinary glucose level (6:00-24:00) was not significantly different between groups; Urinary frequency (0:00-6:00, 6:00-24:00, 24 h) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p<0.0001, p=0.03, p<0.0001, respectively); Urinary volume (0:00-6:00) was significantly lower in patients on tofogliflozin than in those on ipragliflozin (p=0.0007) (Table 2).

FPG levels and insulin doses on the last day of the first intervention were 85.0 (82.3-89.0) mg/dL, 21.5 (14.0-30.0) U/day, respectively. Those on the first day of the second intervention were 91.0 (82.3-100.5) mg/dL, 21.0 (12.5-29.5) U/day, respectively.

Table 3 shows parameters of Table 2 in T/I group and I/T group.

In T/I group, FPG levels and insulin doses on the last day of the first intervention were 87.0 (81.0-89.0) mg/dL, 22.0 (12.0-30.0) U/day, respectively. Those on the first day of the second intervention were 92.0 (84.0-98.0) mg/dL, 24.0 (10.0-29.0) U/day, respectively. In I/T group, FPG levels and insulin doses on the last day of the first intervention were 85.0 (83.0-89.0) mg/dL, 21.0 (16.0-28.0) U/day, respectively. Those on the first day of the second intervention were 88.0 (82.5-100.0) mg/dL, 20.0 (16.0-28.0) U/day, respectively.

In patients on ipragliflozin, 21 (70%) patients had nocturnal hypoglycemia. We investigated the relationship between the risk of nocturnal hypoglycemia and BMI in patients on ipragliflozin. Low BMI was significantly associated with increased nocturnal hypoglycemia (odds ratio 0.51, 95% confidence interval [CI] 0.29-0.88; p=0.02: logistic regression analysis).

Table 2 Parameters of glycemic variability and urinary (and basal insulin dose) in patients treated with tofogliflozin 20 mg + insulin glargine 300 U/mL or ipragliflozin 50 mg + insulin glargine 300 U/mL

	Tofogliflozin 20 mg	Ipragliflozin 50 mg	*p*
0:00 to 6:00 area over the glucose curve (AOC) (<70 mg/dL), mg·min/dL	**0 (0-0)**	**162.2 (0-1,698.8)**	**0.001**
24 h AOC (<70 mg/dL), mg·min/dL	**0 (0-0)**	**329.6 (0-2,596.9)**	**0.002**
8:00 to 24:00 AOC (<70 mg/dL), mg·min/dL	0 (0-0)	0 (0-0)	0.35
24 h M-value (target glucose level = 100 mg/dL)	7.3 (4.0-14.6)	11.4 (5.1-20.6)	0.008
0:00 to 6:00 M-value (target glucose level = 90 mg/dL)	**0.4 (0.1-1.0)**	**3.1 (1.3-5.4)**	**<0.0001**
8:00 to 24:00 M-value (target glucose level = 120 mg/dL)	3.8 (1.7-7.3)	6.6 (2.4-15.0)	0.0003
Mean amplitude of glycemic excursion (MAGE), mg/dL	**51.1 (36.7-74.6)**	**71.9 (48.0-87.2)**	**0.0003**
Mean of daily difference (MODD), mg/dL	21.5 (16.9-28.1)	22.9 (15.8-31.6)	0.57
Average daily risk range (ADRR)	**10.3 (5.9-20.0)**	**22.0 (9.9-29.3)**	**0.001**
24 h mean glucose level, mg/dL	132.8 (118.0-149.8)	141.7 (120.0-159.6)	0.009
0:00 to 6:00 mean glucose level, mg/dL	92.2 (84.6-98.2)	96.4 (72.2-118.3)	0.86
8:00 to 24:00 mean glucose level, mg/dL	**147.3 (128.3-167.8)**	**157.9 (139.8-194.0)**	**0.0001**
24 h standard deviation (SD), mg/dL	35.9 (23.6-54.1)	50.2 (31.7-62.5)	0.0002
0:00 to 6:00 SD, mg/dL	**6.3 (4.6-11.3)**	**12.3 (9.2-17.6)**	**0.0007**
8:00 to 24:00 SD, mg/dL	**31.1 (20.0-40.9)**	**42.7 (24.8-52.8)**	**0.0001**
Area under the glucose curve (AUC) (≥140 mg/dL) within 3 hours after each meal, mg·min/dL			
Breakfast	6,648.8 (2,796.9-10,985.3)	8,983.1 (3,859.4-19,010.9)	0.02
Lunch	4,841.3 (174.1-16,889.4)	10,272.5 (3,254.4-23,915.0)	0.003
Supper	5,520.8 (1,643.3-12,327.9)	12,574.4 (4,997.8-23,417.1)	0.0003
24 h AUC (≥0 mg/dL), mg·min/dL	191,275.8 (169,952.7-215,651.3)	202,345.5 (172,852.3-230,015.2)	0.007
Basal insulin dose, U/day	20.5 (12.0-30.0)	21.5 (14.0-30.8)	0.16
0:00 to 6:00 urinary glucose level, g/day	2.9 (1.1-5.8)	6.8 (3.0-9.8)	0.0002
6:00 to 24:00 urinary glucose level, g/day	27.1 (16.3-41.0)	26.9 (15.9-40.1)	0.7
24 h urinary glucose level, g/day	30.3 (20.7-45.0)	34.5 (20.7-49.8)	0.03
0:00 to 6:00 urinary frequency, times/day	1.0 (1.0-2.0)	2.0 (1.25-3.0)	<0.0001
6:00 to 24:00 urinary frequency, times/day	6.0 (5.0-7.0)	7.0 (5.0-8.0)	0.03
24 h urinary frequency, times/day	7.0 (5.3-8.8)	9.0 (7.0-10.8)	<0.0001
0:00 to 6:00 urinary volume, mL/day	245.0 (102.5-400.0)	500.0 (350.0-500.0)	0.0007
6:00 to 24:00 urinary volume, mL/day	1,175.0 (812.5-1,537.5)	1,110.0 (912.5-1,500.0)	0.73
24 h urinary volume, mL/day	1,400.0 (1,112.5-1,800.0)	1,635.0 (1,255.0-2,000.0)	0.08

Data are shown as median (interquartile range). *p*: Wilcoxon signed-rank test. Primary endpoint parameters are represented by bold font.

Table 3 Parameters of Table 2 in T/I group and I/T group

	T/I group			I/T group		
	Tofogliflozin 20 mg	Ipragliflozin 50 mg	*p*	Tofogliflozin 20 mg	Ipragliflozin 50 mg	*p*
0:00 to 6:00 area over the glucose curve (AOC) (<70 mg/dL), mg·min/dL	**0 (0-0)**	**300.0 (0-1,931.3)**	**0.09**	**0 (0-0)**	**129.3 (3.8-1,545.0)**	**0.004**
24 h AOC (<70 mg/dL), mg·min/dL	**0 (0-30)**	**359.3 (0-2,145.0)**	**0.09**	**0 (0-0)**	**300.0 (3.8-3,210.0)**	**0.004**
8:00 to 24:00 AOC (<70 mg/dL), mg·min/dL	0 (0-0)	0 (0-0)	0.32	0 (0-0)	0 (0-0)	0.47
24 h M-value (target glucose level = 100 mg/dL)	9.8 (5.8-17.2)	10.7 (4.2-21.9)	0.28	6.1 (2.5-11.1)	12.1 (5.5-18.8)	0.005
0:00 to 6:00 M-value (target glucose level = 90 mg/dL)	**0.5 (0.1-1.3)**	**2.0 (1.0-4.4)**	**0.001**	**0.3 (0.1-0.7)**	**4.4 (2.8-5.4)**	**0.001**
8:00 to 24:00 M-value (target glucose level = 120 mg/dL)	5.3 (2.4-11.3)	6.1 (2.8-14.9)	0.04	2.6 (1.0-5.9)	9.1 (2.2-14.8)	0.002
Mean amplitude of glycemic excursion (MAGE), mg/dL	**55.4 (38.3-82.7)**	**72.2 (48.6-91.2)**	**0.02**	**51.0 (33.1-64.9)**	**71.6 (44.7-86.8)**	**0.006**
Mean of daily difference (MODD), mg/dL	21.7 (19.3-26.2)	18.8 (17.2-29.2)	0.73	20.7 (15.8-29.6)	23.3 (15.3-31.4)	0.26
Average daily risk range (ADRR)	**10.8 (6.9-16.2)**	**26.0 (11.2-28.7)**	**0.04**	**9.2 (5.5-21.3)**	**21.4 (9.2-29.4)**	**0.009**
24 h mean glucose level, mg/dL	137.2 (129.0-159.2)	138.2 (130.6-153.2)	0.5	120.6 (108.4-140.8)	144.1 (119.3-165.9)	0.005
0:00 to 6:00 mean glucose level, mg/dL	94.9 (84.8-101.9)	95.3 (73.2-115.0)	0.65	92.1 (85.5-95.5)	97.0 (75.8-119.4)	0.5
8:00 to 24:00 mean glucose level, mg/dL	**153.0 (144.4-174.9)**	**157.0 (144.3-191.6)**	**0.06**	**141.4 (125.5-159.5)**	**173.5 (140.3-192.3)**	**0.0007**
24 h standard deviation (SD), mg/dL	35.0 (25.0-58.9)	50.3 (33.4-67.8)	0.009	36.8 (21.8-45.5)	50.0 (30.4-56.8)	0.006
0:00 to 6:00 SD, mg/dL	**5.7 (4.5-8.1)**	**13.1 (10.6-18.5)**	**0.003**	**8.9 (5.0-12.3)**	**9.4 (7.8-13.2)**	**0.13**
8:00 to 24:00 SD, mg/dL	**37.6 (21.6-47.6)**	**45.0 (26.8-51.5)**	**0.009**	**25.0 (16.4-39.7)**	**42.5 (23.0-52.4)**	**0.005**
Area under the glucose curve (AUC) (≥140 mg/dL) within 3 hours after each meal, mg·min/dL						
Breakfast	9,436.3 (4,365.6-17,183.8)	10,125.0 (4,791.3-19,475.0)	0.11	5,385.0 (408.8-8,377.5)	6,598.8 (1,685.0-17,868.1)	0.046
Lunch	11,431.3 (2,831.3-15,848.7)	11,500.0 (4,793.3-23,492.5)	0.05	517.5 (0-13,174.4)	7,131.3 (2,003.1-18,760.0)	0.02
Supper	10,412.5 (3,698.8-15,230.6)	13,298.8 (5,028.1-24,641.9)	0.02	5,061.7 (6.4-8,938.8)	11,850.0 (5,919.8-19,151.4)	0.005
24 h AUC (≥0 mg/dL), mg·min/dL	197,988.5 (185,784.0-229,046.6)	198,790.0 (188,139.7-220,671.9)	0.53	173,634.7 (156,112.0-202,726.8)	202,345.5 (171,776.9-239,016.7)	0.003
Basal insulin dose, U/day	22.0 (12.0-30.0)	24.0 (8.5-30.5)	0.53	18.0 (13.5-26.5)	21.0 (16.0-28.0)	0.17
0:00 to 6:00 urinary glucose level, g/day	3.1 (1.7-5.8)	6.2 (3.4-9.3)	0.03	2.5 (0.5-6.5)	7.4 (2.4-10.4)	0.001
6:00 to 24:00 urinary glucose level, g/day	25.8 (19.4-40.0)	26.6 (21.3-34.2)	0.61	30.1 (15.2-41.4)	30.8 (13.7-42.4)	0.75
24 h urinary glucose level, g/day	30.2 (24.7-42.3)	34.3 (28.2-40.9)	0.51	33.4 (18.0-43.7)	40.2 (17.7-51.2)	0.02
0:00 to 6:00 urinary frequency, times/day	1.0 (1.0-2.0)	2.0 (2.0-3.0)	0.003	1.0 (0.5-2.0)	2.0 (1.0-2.0)	0.008
6:00 to 24:00 urinary frequency, times/day	6.0 (5.0-7.0)	6.0 (5.0-9.0)	0.07	5.0 (4.5-7.0)	7.0 (5.5-7.5)	0.15
24 h urinary frequency, times/day	7.0 (7.0-8.5)	9.0 (7.0-10.5)	0.003	7.0 (5.0-8.5)	8.0 (7.5-10.0)	0.007
0:00 to 6:00 urinary volume, mL/day	200.0 (125.0-395.0)	500.0 (375.0-500.0)	0.008	300.0 (50.0-500.0)	500.0 (340.0-500.0)	0.02
6:00 to 24:00 urinary volume, mL/day	1,200.0 (900.0-1,775.0)	1,200.0 (1,060.0-1,550.0)	0.65	1,100.0 (725.0-1,250.0)	1,100.0 (770.0-1,425.0)	0.88
24 h urinary volume, mL/day	1,400.0 (1,200.0-1,975.0)	1,700.0 (1,560.0-2,000.0)	0.2	1,400.0 (1,050.0-1,800.0)	1,500.0 (1,155.0-2,000.0)	0.17

Data are shown as median (interquartile range). *p*: Wilcoxon signed-rank test. Primary endpoint parameters are represented by bold font.

We determined cut-off value of BMI, which has the highest prediction ability for nocturnal hypoglycemia, by using receiver operating characteristic analysis. When the cutoff value was 24.4 kg/m^2, which has the highest prediction ability, the sensitivity was 81% and the specificity was 89%. The area under the curve for nocturnal hypoglycemia was 0.93 (95% CI, 0.83-1.00; p<0.0001) (Fig. 3).

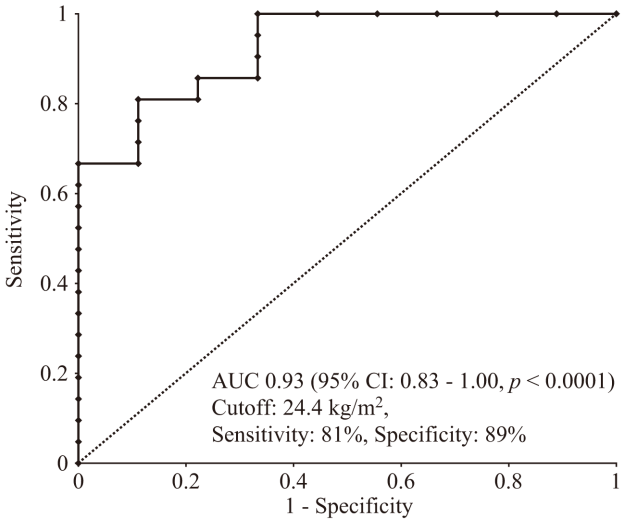

Fig. 3 A Receiver Operating Characteristic (ROC) curve for nocturnal hypoglycemia in Body Mass Index (BMI)
When the cutoff value was 24.4 kg/m^2, which has the highest prediction ability, the sensitivity was 81% and the specificity was 89%. The area under the curve for nocturnal hypoglycemia was 0.93 (95% CI, 0.83-1.00; p<0.0001).

Discussion

The results of this study suggest that tofogliflozin, which reduces glycemic variability by avoiding nocturnal hypoglycemia and decreasing daytime (postprandial) glucose levels, is an ideal SGLT2i when used together with Glargine 300, during basal insulin therapy.

In this study, FPG levels were titrated to 80 mg/dL level by Glargine 300. In this case, if we think that nocturnal glucose levels are affected only by Glargine 300, nocturnal glucose levels are thought to be theoretically titrated to almost 80 mg/dL level. Tofogliflozin has a short half-life, and when administered in the morning, its effect almost disappears by nighttime [12]. Therefore, further decreases in nocturnal glucose levels are not thought to be caused due to intake of tofogliflozin. In contrast, ipragliflozin is thought to pose a risk of further decrease in nocturnal glucose levels, in combination with insulin because ipragliflozin has a long half-life, and therefore acts firmly during the nighttime [22]. We think that occurrence of nocturnal and overall hypoglycemia was significantly lower in patients on tofogliflozin, than in those on ipragliflozin because of the above-mentioned difference between tofogliflozin and ipragliflozin.

In this study, the mean glucose level (0:00-6:00) was not significantly different between patients on tofogliflozin and ipragliflozin (this was expected, because FPG levels were titrated at 80 mg/dL level). M value (0:00-6:00) was significantly lower in patients on tofogliflozin, than in those on ipragliflozin. This was thought to occur because nocturnal glucose levels came closer to the target glucose levels (90 mg/dL) in patients on tofogliflozin, than in those on ipragliflozin, since occurrence of nocturnal hypoglycemia was significantly lower in patients on tofogliflozin, than in those on ipragliflozin.

In this study, mean glucose level (8:00-24:00) and SD (8:00-24:00) were significantly lower in patients on tofogliflozin, than in those on ipragliflozin. These were thought to occur because the effect of tofogliflozin is suggested to be stronger during the daytime (and weaker during the nighttime) than that of ipragliflozin even though there are no differences between the overall effects of tofogliflozin or ipragliflozin [10, 11]. The reason of the following is estimated that a half-life of tofogliflozin is shorter than that of ipragliflozin [12, 22]. In contrast, the nocturnal effect of ipragliflozin became less because SGLT2i decreases glucose levels in a glucose level-dependent manner. Therefore, this may have led to mean glucose levels (24 hours) also being significantly lower in patients on tofogliflozin, than in those on ipragliflozin.

In this study, MAGE, ADRR, and SD (24 hours) were significantly lower in patients on tofogliflozin, than in those on ipragliflozin. The effect of tofogliflozin which has a short half-life is suggested to be stronger during the daytime and disappears quickly during nighttime, as compared to that of ipragliflozin, even though there are no differences between the overall effects of tofogliflozin and ipragliflozin [10-12, 22]. Therefore, daytime glucose levels were lower, and occurrence of nocturnal hypoglycemia was lower in patients on tofogliflozin, than in those on ipragliflozin. This leads to reduced glycemic variability overall, and maybe responsible for the results observed in this study.

Further, we believe that tofogliflozin's short half-life led to this study results of urinary endpoint. From the viewpoint of adverse effect, tofogliflozin is superior in reducing urinary frequency. Regarding that urinary glucose level during the daytime in patients on tofogliflozin were almost the same as in those on ipragliflozin and urinary frequency during the daytime was lower in patients on tofogliflozin than in those on ipragliflozin but blood glucose level during the daytime was lower in patients on tofogliflozin than in those on ipragliflozin, SGLT2i decreases blood glucose levels by lowering renal glucose threshold, therefore, there is less necessity for tofogliflozin to excrete urinary glucose during the daytime than ipragliflozin because blood glucose level during the daytime was lower in patients on tofogliflozin than in those on ipragliflozin.

In a report supporting the results of this study, it has been stated that the clinical effect of Glargine 300 lasts for 24 hours (steady state) [23]. Glargine 300 is thought to be useful long-acting insulin to decrease FPG levels, reducing the occurrence of nocturnal hypoglycemia. It has also been reported that time-to-maximum blood concentration (Tmax) of tofogliflozin is 1.1 hours, and the half-life of tofogliflozin is 5.4 hours [12]. Approximately 17 hours after tofogliflozin is taken, only 12.5% of the Tmax remains. Therefore, this supports the hypothesis that the effect of tofogliflozin taken in the morning almost disappears during the nighttime. In contrast, it has been reported that Tmax of ipragliflozin is 1.4 hours and its half-life is 15 hours [22]. Therefore, ipragliflozin affects the individual for a constant duration (steady state). Considering together with the previous reports [10, 11], this supports the hypothesis that the effect of ipragliflozin persists during the nighttime, and that it does not have a strong effect only during the daytime.

It has been reported that the risk factors for hypoglycemia are low BMI and concomitant insulin use in the elderly subjects with type 2 diabetes treated with ipragliflozin [24]. We might also investigate the relationship between nocturnal hypoglycemia and BMI in patients on ipragliflozin, therefore, we didn't set a limit of BMI in selection criteria of study subjects. The present study results suggest that patients who aren't obecity should avoid taking ipragliflozin and should take tofogliflozin especially.

Clarifying role-sharing of supporting basal and bolus insulin secretion is important in order to clarify an approach of basal insulin therapy, and to realize ideal glycemic variability during basal insulin therapy. We examine both these therapies in this study. In the combination therapy consisting of Glargine 300 + tofogliflozin, Glargine 300 supplies basal insulin and tofogliflozin supports bolus insulin secretion. Thus, the role-sharing of "basal + bolus" in this combination is clearly defined. In contrast, in the combination therapy consisting of Glargine 300 + ipragliflozin, Glargine 300 supplies basal insulin and ipragliflozin supports basal and bolus insulin secretion. Thus, role-sharing in this case "basal + (basal + bolus)" is not distinct. It is thought to be ideal if supply of basal insulin is left to Glargine 300, and effects of OHA as well as supplying of bolus insulin are integrated into one drug, such as tofogliflozin. In basal insulin therapy, supporting basal insulin secretion by OHA leads to loss of efficacy of the OHA which decreases glucose levels in a glucose level-dependent manner, and carries a very high risk of nocturnal hypoglycemia in the case of OHA which decreases glucose levels in a glucose level-independent manner. In the case of SGLT2i, the effect is lost because SGLT2i decreases glucose levels in a glucose level-dependent manner, and risk of hypoglycemia increases in combination with insulin. This is inefficient and increases risk to the patient; therefore, this combination is undesirable. In contrast, tofogliflozin administered in the morning is efficient because tofogliflozin almost affects to support bolus insulin secretion and reduces the risk of nocturnal hypoglycemia in combination with insulin, because its effect is stronger during the daytime and disappears during the nighttime. Therefore, we believe that tofogliflozin which is efficient and reduces risk of nocturnal hypoglycemia is an ideal SGLT2i.

The results of this study suggest that tofogliflozin, which reduces glycemic variability by preventing nocturnal hypoglycemia and decreasing daytime (postprandial) glucose levels, is superior when used together with Glargine 300 for basal insulin therapy, compared to ipragliflozin. The combination therapy of Glargine 300 + tofogliflozin may be the ideal therapy, since it improves glycemic variability by supporting basal and bolus insulin secretion accurately, reducing the risk of nocturnal hypoglycemia maximally, during basal insulin therapy in which the patient's preferences and view-point are considered. Thus, the clinical significance of this study is high. However, the present study was limited by certain factors. First, it is a single facility, open-label study; second, this study

is demonstrated in pretty much short time; third, the PK/PD comparative data between the action of tofogliflozin and that of ipragliflozin is not exist. We shall endeavor to address these limitations by gathering more cases and conducting further clinical studies in the future.

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S.T. (corresponding author) designed the study, collected and analyzed the data, wrote and reviewed the manuscript, and revised it critically for important intellectual content. H.T. and S.T. contributed to the review of the manuscript. S.T. (corresponding author) is the guarantor of this work and, as such, had full access to all the data in the study, and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Disclosure

None of the authors have any relevant conflicts of interest to disclose.

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