Factors with remission of fatty liver in patients with type 2 diabetes treated with ipragliflozin

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Abstract. We investigated the factors associated with fatty liver remission via treatment with ipragliflozin. The analysis was obtained from our multi-center prospective observational study, including 200 Japanese patients with type 2 diabetes treated with ipragliflozin (50 mg/day) for 24 weeks. The extent of fatty liver was estimated using a fatty liver index (FLI). Based on the FLI after the treatment with ipragliflozin, patients were classified into remission group (FLI < 30) and non-remission group (FLI ≥ 30). After treatment with ipragliflozin for 24 weeks, FLI significantly improved from 64.5 ± 21.6 to 51.9 ± 26.5 (p < 0.01). Body weight, body mass index, waist circumference, aspartate aminotransferase, alanine aminotransferase, and FLI in the remission group were significantly lower compared with those of the non-remission group. Stepwise analysis showed that the baseline FLI (Odds ratio 0.86; 95% confidence interval 0.81–0.90, p < 0.01) was an independent factor associated with FLI remission. Using a receiver operating characteristic (ROC) analysis, the adequate cut-off value for the remission was 50. The area under the ROC curve was 0.93 with the sensitivity and specificity 84.6% and 90.1% respectively.

In conclusion, ipragliflozin ameliorated fatty liver. These results suggest that patients with fatty liver with a lower FLI are more likely to attain remission by the treatment with ipragliflozin.

Key words: Fatty liver, Ipragliflozin, Diabetes

FATTY LIVER DISEASE AND TYPE 2 DIABETES

often coexist, sharing common pathogenic factors such as insulin resistance [1]. Fatty liver disease, including steatosis, hepatic cirrhosis, and hepatocellular carcinoma, is a major cause of mortality [2]. Although fatty liver disease related to diabetes is increasing [3, 4], the treatment is not adequate. Previous reports have shown that ipragliflozin, a sodium-glucose cotransporter 2 (SGLT2) inhibitor, improved hepatic steatosis in rodent models [5-7]. Moreover, in our preliminary clinical study, ipragliflozin had favorable effects for fatty liver in a small number of patients with type 2 diabetes [8]. In the current study, we investigated whether ipragliflozin ameliorated fatty liver in a larger sample size. In addition, the factors associated with fatty liver remission via treatment with ipragliflozin were explored.

Materials and Methods

This sub-analyzed data were obtained from our multi-center prospective observational study [9]. The study included 200 Japanese patients (56 women) with type 2 diabetes who were undergoing outpatient treatment from February 2015 to July 2017. The inclusion criteria were as follows: aged between 20 and 75, HbA1c level between 6.5% and 8.5%, body mass index (BMI) above 22 kg/m², estimated glomerular filtration rate (eGFR) above 45 mL/min/1.73 m², and receipt of sulfonylurea therapy for at least 12 weeks prior to enrolment. After obtaining patient’s informed consent, once-daily ipragliflozin (50 mg/day) was given for 24 weeks. Participants either remained on sulfonylurea, but reduced to the lowest dose, or discontinued sulfonylurea according to the physician’s decision. Several clinical parameters, including fasting plasma glucose (FPG), HbA1c, lipid profiles and liver function were compared before and after 24
weeks of treatment. The study was registered with the University Hospital Medical Information Network (UMIN) Center (#000016347) prior to enrolment, approved by the Institutional Review Board of Hokkaido Hospital.

The extent of fatty liver was estimated using a fatty liver index (FLI), comprising BMI, waist circumference (WC), gamma-glutamyl transferase (γ-GTP) and triglycerides (TG). FLI was calculated using the following equation: 

\[ \text{FLI} = \frac{(\exp(0.953 \times \log(TG) + 0.139 \times \text{BMI} + 0.718 \times \log(\text{GGT}) + 0.053 \times \text{WC} - 15.745))/\left(1 + \exp(0.953 \times \log(TG) + 0.139 \times \text{BMI} + 0.718 \times \log(\text{GGT}) + 0.053 \times \text{WC} - 15.745)\right)}{100} \]  

Because FLI < 30 is used to rule out hepatic steatosis [11, 12], patients with FLI < 30 were excluded. Based on the FLI after 24 weeks of treatment with ipragliflozin, these patients were classified into the remission group (FLI < 30) and the non-remission group (FLI ≥ 30).

Results were expressed as mean ± standard deviation or median. Differences in baseline characteristics and changes between the groups were compared using the unpaired t-test, the chi-square test or Mann-Whitney U-test, as appropriated. Differences of clinical parameters before and after 24 weeks of treatment were analyzed using a paired t-test. Simple linear regression analyses were performed to test for associations between variables. Multivariate analyses were performed using stepwise regression to identify factors independently associated with the outcomes. A receiver operating characteristic (ROC) curve was analyzed to define the cut-off values for fatty liver remission. All p values were two-sided and values <0.05 were considered statistically significant. Statistical analysis was performed using JMP Pro version 14 (SAS Inc., Cary, NC, USA).

Results

Of the 200 patients, 50 were not analyzed due to the exclusion criteria of FLI less than 30 (n = 34) or missing data (n = 16). Consequently, 150 patients (39 women) were included in the analysis. 39 patients were in remission group, and 111 patients in non-remission group (Fig. 1). At baseline, the mean age and HbA1c level in the whole patients were 56.9 ± 9.6 years and 7.6 ± 0.5%, respectively. After 24 weeks, FLI significantly improved from 64.5 ± 21.6 to 51.9 ± 26.5 (p < 0.01). The changes in FLI showed a significant positive correlation with the changes in FPG (Correlation coefficient = 0.22, p < 0.01, Supplementary Fig. 1). No significant changes were shown in FIB-4 index, one of a liver fibrosis marker (data not shown).

The clinical and biochemical characteristics of the patients are shown in Table 1. Body weight, BMI, WC, aspartate aminotransferase (AST), alanine aminotransferase (ALT), and FLI in the remission group were significantly lower compared with those of the non-remission group. The proportion of the patients who remained on sulfonylurea in the remission group was significantly higher than those in the non-remission group (64.1% vs. 41.4%, p < 0.05), but there were no differences in FPG nor HbA1c between the two groups. Similar results were observed in the clinical and biochemical characteristics after treatment with ipragliflozin (Supplementary Table 1).

Stepwise analysis was performed with parameters showing statistical significant differences at baseline. The result identified that the baseline FLI was an independent factor associated with the remission (Odds ratio 0.85; confidence interval 0.81–0.90, p < 0.01). Using a receiver operating characteristic (ROC) analysis, the adequate cut-off value for the remission was 50. The area under the ROC curve was 0.93 with the sensitivity and specificity 84.6% and 90.1% respectively (Fig. 2). In addition, we classified patients into low FLI group (30 ≤ FLI < 62, n = 75) and high FLI group (62 ≤ FLI < 100, n = 75), and examined association with changes in FLI. FLI significantly improved in patients with low FLI at baseline (–16 (–9, –21) vs. –9 (–3, –19), p < 0.05, Fig. 3).

Discussion

This study showed that ipragliflozin ameliorated FLI, used as a surrogate measure for fatty liver, with a larger sample size than our previous study. This result was consistent with a post-marketing surveillance study [13]. The changes in FLI had significant positive correlations with those in FPG, in consistent with our previous report [8] as well. Moreover, our results indicated that low FLI
Before treatment was an independent factor to predict fatty liver remission via treatment with ipragliflozin.

FLI significantly improved in patients with low FLI at baseline. Patients with low FLI are mildly obese in this study. FLI also improved in patients with low BMI at baseline (Correlation coefficient = 0.28, \( p < 0.01 \), Supplementary Fig. 2). The relationship between mild obesity at baseline and the fatty liver remission may be explained by the effect of ipragliflozin on adipocyte. Fatty liver is a marker for the failure of the adipocyte to expand to accommodate an increased energy influx [14]. Japanese have reduced adipocyte capacity, and tend to become fatty liver with a small increase in BMI [14]. It has been reported that ipragliflozin promoted normotopic fat accumulation and prevented ectopic fat accumulation in the liver in rodent models [7]. Taken together, fatty liver remission could be more pronounced by improving adipocyte capacity to store fat in mild obese patients with a low FLI by the treatment with ipragliflozin.

The limitation of this study is that all patients were received sulfonylurea therapy for at least 12 weeks prior to enrolment. Thus, the influence of discontinued sulfonylurea therapy was unclear before treatment.

### Table 1

|                      | Remission group \((n = 39)\) | Non-remission group \((n = 111)\) | \( p \) value |
|----------------------|-------------------------------|----------------------------------|---------------|
| Age                  | 61.9 ± 7.3                    | 55.1 ± 9.7                       | <0.01         |
| Sex (woman %)        | 18.0                          | 28.8                             | 0.18          |
| Body weight (kg)     | 69.6 ± 8.2                    | 83.7 ± 15.7                     | <0.01         |
| BMI (kg/m\(^2\))     | 25.7 ± 2.0                    | 30.5 ± 4.8                      | <0.01         |
| Waist circumference (cm) | 90.6 ± 5.7                    | 101.3 ± 10.4                    | <0.01         |
| FPG (mg/dL)          | 145.6 ± 38.8                  | 153.6 ± 36.9                    | 0.25          |
| HbA1c (%)            | 7.5 ± 0.5                     | 7.6 ± 0.5                       | 0.20          |
| AST (U/L)            | 22 (19–28)                    | 29 (22–40)                      | <0.01         |
| ALT (U/L)            | 25 (22–40)                    | 37 (24–56)                      | <0.01         |
| γ-GTP (U/L)          | 30 (22–38)                    | 46 (29–79)                      | <0.01         |
| eGFR (ml/min/1.73 m\(^2\)) | 77.3 ± 17.7                  | 77.8 ± 19.2                     | 0.89          |
| TG (mg/dL)           | 114 (93–163)                  | 132 (94–185)                    | 0.20          |
| T-Chol (mg/dL)       | 173.0 ± 23.8                  | 184.0 ± 32.0                    | 0.06          |
| HDL-C (mg/dL)        | 50.7 ± 12.9                   | 49.0 ± 12.2                     | 0.48          |
| LDL-C (mg/dL)        | 97.0 ± 21.9                   | 103.8 ± 29.3                    | 0.20          |
| non-HDL-C (mg/dL)    | 122.7 ± 22.3                  | 135.1 ± 30.9                    | <0.05         |
| SU (low dose group %)| 64.1                          | 41.4                             | <0.05         |
| FLI                  | 40.9 ± 10.3                   | 72.7 ± 18.2                     | <0.01         |

Data are expressed as mean ± standard deviation or median. BMI, body mass index; FPG, fasting plasma glucose; HbA1c, hemoglobin A1c; AST, aspartate amino transferase; ALT, alanine amino transferase; γ-GTP, gamma-glutamyl transferase; eGFR, estimated glomerular filtration rate; TG, triglycerides; T-Chol, total-cholesterol; HDL-C, high density lipoprotein cholesterol; LDL-C, low density lipoprotein cholesterol; non-HDL-C, non-high density lipoprotein cholesterol; SU, sulfonylurea; FLI, fatty liver index.

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**Fig. 2** Receiver operating characteristic curves analysis for identifying the cutoff value of fatty liver index (FLI) for determining fatty liver remission.
nylurea cannot be excluded. In addition, since fatty liver was estimated indirectly by calculating FLI, pathological examination or imaging are needed to validate the effect of ipragliflozin on fatty liver in a placebo-controlled randomized clinical trial.

In conclusion, ipragliflozin ameliorated fatty liver. This result suggests that patients with a fatty liver with a lower FLI are more likely to attain remission by the treatment with ipragliflozin.

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Disclosure

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### Supplementary Table 1  The clinical and biochemical characteristics of the patients after the treatment with ipragliflozin

|                          | Remission group (n = 39) | Non-remission group (n = 111) | p value |
|--------------------------|--------------------------|-------------------------------|---------|
| Body weight (kg)         | 65.6 ± 7.5               | 80.0 ± 15.2                   | <0.01   |
| BMI (kg/m²)              | 24.2 ± 1.7               | 29.2 ± 4.6                    | <0.01   |
| Waist circumference (cm) | 86.3 ± 5.2               | 97.3 ± 9.6                    | <0.01   |
| FPG (mg/dL)              | 132.6 ± 19.2             | 139.4 ± 29.2                  | 0.17    |
| HbA1c (%)                | 7.2 ± 0.5                | 7.4 ± 0.6                     | 0.06    |
| AST (U/L)                | 19 (17–23)               | 23 (18–29)                    | <0.01   |
| ALT (U/L)                | 17 (14–25)               | 26 (21–38)                    | <0.01   |
| γ-GTP (U/L)              | 21 (19–27)               | 37 (23–55)                    | <0.01   |
| eGFR (mL/min/1.73 m²)    | 79.4 ± 18.6              | 79.6 ± 19.4                   | 0.94    |
| TG (mg/dL)               | 87 (62–115)              | 134 (96–198)                  | <0.01   |
| T-Cho (mg/dL)            | 172.4 ± 26.9             | 188.4 ± 32.1                  | <0.01   |
| HDL-C (mg/dL)            | 57.7 ± 14.8              | 52.2 ± 13.4                   | <0.05   |
| LDL-C (mg/dL)            | 96.2 ± 22.0              | 104.0 ± 28.1                  | 0.12    |
| non-HDL-C (mg/dL)        | 114.7 ± 24.4             | 136.1 ± 31.5                  | <0.01   |
| FLI                      | 20.8 ± 4.6               | 62.8 ± 21.9                   | <0.01   |

Data are expressed as mean ± standard deviation or median. BMI, body mass index; FPG, fasting plasma glucose; HbA1c, hemoglobin A1c; AST, aspartate amino transferase; ALT, alanine amino transferase; γ-GTP, gamma-glutamyl transferase; eGFR, estimated glomerular filtration rate; TG, triglycerides; T-Cho, total-cholesterol; HDL-C, high density lipoprotein cholesterol; LDL-C, low density lipoprotein cholesterol; non-HDL-C, non-high density lipoprotein cholesterol; FLI, fatty liver index.

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### Supplementary Fig. 1

Relationship between the changes in fasting plasma glucose (Δ FPG) and the changes in fatty liver index (Δ FLI) before and 24 weeks after treatment with ipragliflozin

### Supplementary Fig. 2

Relationship between the body mass index (BMI) at baseline and the changes in fatty liver index (Δ FLI) before and 24 weeks after treatment with ipragliflozin

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