Real-world risk of lower-limb amputation associated with sodium–glucose cotransporter 2 inhibitors versus metformin: A propensity score-matched model analysis in Japan

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INTRODUCTION
Type 2 diabetes is a metabolic disorder characterized by elevated levels of plasma glucose. Hyperglycemia is related to an increase in insulin resistance and a decrease in β-cell function, and long-standing hyperglycemia can cause microvascular and macrovascular complications. Lower-limb amputation is a serious outcome in patients with type 2 diabetes. In particular, amputation risk is reportedly increased in patients with poorly controlled and long-standing type 2 diabetes, especially in those requiring insulin therapy. In Japan, the amputation rate in patients with diabetes and peripheral artery disease (PAD) was reportedly 4.43-fold higher than that in patients without diabetes having PAD.
Currently, sodium–glucose cotransporter 2 inhibitors (SGLT2is) are popularly prescribed to patients with type 2 diabetes. SGLT2is ameliorate hyperglycemia through inhibiting the renal proximal tubular reabsorption of glucose, independently of insulin secretion. This distinct class of oral hypoglycemic agents can reduce glycated hemoglobin levels by 0.6–0.8% without increasing the risk of hypoglycemia, thereby increasing time in range, reducing bodyweight and improving metabolic factors, including blood pressure and lipid profile. In contrast, the CANAgli program showed an approximately twofold incidence of lower-limb amputation in patients with type 2 diabetes treated with canagliflozin than that in those treated with placebo (6.3 vs 3.4 participants per 1,000 patient-years).6

The mechanism for the risk of SGLT2is-associated amputation is unclear; this adverse event might be associated with volume depletion, leading to circulation insufficiency in the peripheral vasculature, raising concerns about the risk of amputations associated with canagliflozin and other SGLT2is. Systematic reviews and meta-analyses using randomized controlled trials or observational studies have shown that SGLT2is are not related to the increased risk of amputation. However, whether the findings are true in real-world settings in Japan or not remains unclear.

Thus, the present study aimed to show the actual risk of lower-limb amputation in patients with type 2 diabetes using SGLT2is compared with that in those using metformin as a control in the real-world use of SGLT2is in Japan, and to define the association between amputation risk and patients’ clinical characteristics.

MATERIALS AND METHODS

Study design
This was a retrospective cohort study carried out utilizing the Medical Data Vision (MDV) administrative claims database, a nationwide database using data from Japanese acute care hospitals utilizing the Diagnosis Procedure Combination (DPC) system. The date when SGLT2is or metformin was first prescribed to patients in the SGLT2is and metformin groups, respectively, was defined as the observation start date. Patients were censored at the timing of undergoing lower-limb amputation; discontinuation of target drugs (SGLT2is or metformin); addition of metformin or SGLT2is administration in patients in the SGLT2is or metformin groups, respectively; loss of insurance coverage; or death. The minimum observation period was 1 day after the initiation of SGLT2is or metformin.

Study population
First, we extracted data of 3,129,105 patients with diabetes mellitus (International Classification of Diseases, 10th revision [ICD-10] code: E11–E14) registered between 1 April 2008 and 31 October 2019 in the MDV database (Figure 1). Next, we identified patients with type 2 diabetes aged ≥18 years who were newly prescribed SGLT2is and/or metformin after April 2014, the month in which SGLT2i (ipragliflozin) was available in Japan. The exclusion criteria were as follows: (i) patients diagnosed with type 1 diabetes (ICD-10 code: E10; n = 66,760), those with both type 1 diabetes and type 2 diabetes (ICD-10 code: E11 or E14; n = 179,550), and those with any diabetes mellitus, except type 1 diabetes and type 2 diabetes (n = 860,024); (ii) patients aged <18 years and/or patients with no registration history of SGLT2is and metformin use (n = 1,525,828); and, (iii) patients prescribed metformin before April 2014 (n = 170,778). As a result, 326,165 patients with type 2 diabetes were eligible for inclusion and were subsequently divided into two groups: (i) new users of SGLT2is without preceding metformin use (n = 109,660); and (ii) new users of metformin without preceding SGLT2is use (n = 216,505). Next, we carried out propensity score matching to balance the baseline characteristics (age, sex, body mass index [BMI], and use of concomitant drugs including antidiabetics, antiplatelets, anticoagulants and statins) between the two groups in a 1:1 ratio for SGLT2is to metformin. Finally, a total of 107,296 patients, comprising new users of SGLT2is (n = 53,648) and metformin (n = 53,648), were eligible for inclusion.

Identification of lower-limb amputation events and use of concomitant drugs
Lower-limb amputation events were identified using the relevant ICD-10 codes; that is, Z89.4, Z89.5 and Z89.6. Concomitant drugs were those confirmed to have been prescribed for at least 30 days before SGLT2is or metformin initiation. Atorvastatin, pitavastatin and rosuvastatin were classified as strong statins, whereas other statins were classified as standard statins in the present study.

Outcome
The major outcome was to compare the risks of lower-limb amputation between patients with type 2 diabetes using SGLT2is and metformin. Furthermore, the clinical parameters associated with the increased risk of amputation in patients with type 2 diabetes using SGLT2is and metformin were determined.

Patient characteristics
Age, sex and BMI were identified using data from the claim records within 30 days of the amputation date. Obesity was defined as a BMI of ≥25 kg/m².

Statistical analysis
Normally distributed data (age and BMI) are presented as mean ± standard deviation. Continuous variables were analyzed using the unpaired t-test. Categorical variables were analyzed using the χ²-test or Fisher’s exact test, and are expressed as absolute numbers or percentages. As aforementioned, to minimize or eliminate potential confounding effects, we adjusted for differences in baseline characteristics by 1:1 propensity score matching.
Patients diagnosed with diabetes mellitus 
\( (n = 3,129,105) \)
- Patients diagnosed with T1D \( (n = 66,760) \)
- Patients diagnosed with T1D and T2D \( (n = 179,550) \)
- Patients diagnosed with any other diabetes mellitus except T1D and T2D \( (n = 860,024) \)

Patients diagnosed with T2D 
\( (n = 2,022,771) \)
- Patients aged <18 years and/or patients with no registered history of SGLT2is and/or metformin uses \( (n = 1,525,828) \)
- Patients prescribed before April 2014 \( (n = 170,778) \)

Patients with T2D aged ≥18 years newly prescribed SGLT2is and/or metformin after April 2014 
\( (n = 326,165) \)

New users of SGLT2is without preceding metformin use 
\( (n = 109,660) \)

New users of metformin without preceding SGLT2is use 
\( (n = 216,505) \)

Propensity score estimation and 1:1 matching

Unmatched patients \( (n = 218,869) \)
- New users of SGLT2is \( (n = 56,012) \)
- New users of metformin \( (n = 162,857) \)

- Patients included in the matched cohort \( (n = 107,296) \)
  - New users of SGLT2is eligible for inclusion \( (n = 53,648) \)
  - New users of metformin eligible for inclusion \( (n = 53,648) \)

**Figure 1** | Patient disposition. SGLT2is, sodium–glucose cotransporter 2 inhibitors; T1D, type 1 diabetes; T2D, type 2 diabetes.
matching using the nearest neighbor matching method\(^\text{13}\). After matching, the hazard ratio (HR) for amputation was analyzed using a Cox proportional hazards model. All statistical analyses were carried out using IBM SPSS Statistics for Windows, version 23.0 (IBM Corp., Armonk, NY, USA). Differences were considered statistically significant at a \(P\)-value of <0.05.

**RESULTS**

Table S1 and Table 1 present the baseline clinical characteristics before and after 1:1 propensity score matching, respectively. Before propensity score matching, there were significant differences in many clinical parameters between the SGLT2is and metformin groups, making it difficult to statistically investigate the effect of SGLT2is on amputation (Table S1). After propensity score matching, a total of 107,296 patients (\(n = 53,648\) per group) were selected for the analysis (Table 1).

Lower-limb amputation was carried out in 66 (0.06%) of the 107,296 patients with type 2 diabetes; that is, 25 (0.05%) of 53,648 in the metformin group and 41 (0.08%) of 53,648 in the SGLT2is group (Table 1), over a mean observation period of 142.8 ± 145.5 days. There was no significant difference in the incidence of amputation between the groups, although the incidence tended to be higher in the SGLT2is group than in the metformin group (\(P = 0.065\), \(\chi^2\)-test; Table 1). Furthermore, there were no significant between-group differences in sex, mean age and BMI. In addition, there were no significant differences in the usage rates of all anti-diabetic drugs, excluding thiazolidinediones, antplatelet and anticoagulant drugs, and statins. These findings suggest that most confounding effects could be eliminated, leading to a reliable analysis of the association between SGLT2is use and lower-limb amputation. In the present study, the most frequently used SGLT2i was empagliflozin (30.3%), and the frequencies of the use of the other SGLT2is decreased in the following order: dapaglifoxin (19.4%), irapaglifoxin (18.5%), canagliflozin (15.7%), tofogliflozin (9.6%) and luseogliflozin (6.7%; Table 1).

The clinical characteristics of patients with type 2 diabetes undergoing lower-limb amputation, divided into the metformin and SGLT2is groups, after 1:1 propensity score matching, are presented in Table 2. The mean age and BMI were significantly higher in the SGLT2is group than in the metformin group. In addition, the proportion of obese patients was significantly higher in the SGLT2is group than in the metformin group.

Next, we compared the amputation risk associated with SGLT2is and metformin administration using the Cox proportional hazards model. There was no significant difference in the risk of lower-limb amputation between the SGLT2is and metformin groups, although the risk in the former was seemingly higher than that in the latter (HR 1.34, 95% confidence interval [95% CI] 0.80–2.24; Figure 2). When analyzed according to each SGLT2i drug, there were no significant differences in the risks associated with each SGLT2i drug and metformin.

Subsequently, we compared the risks of lower-limb amputation between the SGLT2is and metformin groups according to the baseline clinical characteristics. The risk was significantly higher in female patients using SGLT2is than in those using metformin (HR 2.78, 95% CI 1.12–6.94, \(P = 0.028\); Figure 3); however, the same finding was not observed in male patients. Regarding age, the risk tended to be higher in patients aged \(\geq 75\) years using SGLT2is than in those using metformin (HR 2.32, 95% CI 0.88–6.13), although the difference was not significant (\(P = 0.090\)). Regarding BMI, although the HR for amputation could not be calculated in type 2 diabetes patients with a BMI of \(\geq 25\) kg/m\(^2\) due to the null event of amputation in those using metformin, the risk of lower-limb amputation was considered higher in patients with BMI of \(\geq 25\) kg/m\(^2\) using SGLT2is than that in those using metformin. Meanwhile, the amputation risk was comparable between type 2 diabetes patients with a BMI of \(<25\) kg/m\(^2\) using SGLT2is and those using metformin. Concomitant anti-diabetic and antplatelet/anticoagulant drugs did not influence amputation risk in the SGLT2is and metformin groups. Regarding statins, amputation risk was significantly higher in strong statin users using SGLT2is than in those using metformin (HR 2.68, 95% CI 1.18–8.20, \(P = 0.046\)).

**DISCUSSION**

The present study showed no significant difference in the risk of lower-limb amputation between patients with type 2 diabetes using SGLT2is and those using metformin. Considering that metformin was not reportedly related to an increased risk of amputation\(^\text{3,14}\), we concluded that SGLT2is were not related to an increased risk of lower-limb amputation in patients with type 2 diabetes. However, a subanalysis according to sex using SGLT2is showed a 2.78-fold increased risk of amputation compared with that using metformin in female patients with type 2 diabetes, but not in male patients. A similar finding was shown in a pooled analysis of the Empagliflozin Cardiovascular Outcome Event Trial in Type 2 Diabetes Mellitus Patients (EMPA-REG OUTCOME), CANVAS Program, Dapagliflozin Effect on Cardiovascular Events—Thrombolysis in Myocardial Infarction 58 (DECLARE-TIMI 58) and Canagliflozin and Renal Events in Diabetes with Established Nephropathy Clinical Evaluation (CREDENCE) trials; that is, the risk ratios for amputation in participants with SGLT2is treatment versus placebo were 1.79 (95% CI 1.08–2.97) and 1.23 (95% CI 0.99–1.52) in women and men, respectively, although there was no statistically significant difference between the two sexes\(^\text{15}\). However, the reason for the increased risk of amputation in women was not discussed and remains unclear. Generally, the hemodynamic effects of diuretic action and blood pressure lowering using SGLT2is are related to an increased risk of amputation\(^\text{16}\). Similar effects were found with diuretic agents, which might decrease peripheral perfusion and subsequently increase the risk of lower-limb amputation in patients with type 2 diabetes\(^\text{16}\). Considering that, in general, the daily intake of water was reportedly lower in women than in men in Japan\(^\text{17}\) and foreign countries\(^\text{18}\), female patients with type 2 diabetes might be more
influenced by the hemodynamic effect of SGLT2is through hypovolemia due to insufficient water intake than male patients; however, this warrants further investigation. Furthermore, the mean age at which amputation was carried out was higher in female patients than in male patients; that is, 66.8 ± 14.5 years and 62.9 ± 12.9 years, respectively, although

| Characteristics                      | Overall (n = 107,296) | Metformin (n = 53,648) | SGLT2is (n = 53,648) | P-value (SGLT2is vs metformin) |
|--------------------------------------|------------------------|------------------------|----------------------|-----------------------------|
| Sex, n (%)                           |                        |                        |                      |                             |
| Male                                 | 70,013 (65.3)          | 35,057 (65.3)          | 34,956 (65.2)        | 0.521†                     |
| Female                               | 37,283 (34.7)          | 18,591 (34.7)          | 18,692 (34.8)        |                            |
| Age Mean (years)                     | 64.4 ± 12.8            | 64.4 ± 12.2            | 64.5 ± 13.4          | 0.227‡                     |
| Distribution, n (%)                  |                        |                        |                      |                             |
| <25                                  | 448 (0.4)              | 209 (0.4)              | 239 (0.4)            | <0.001†                    |
| 25–44                                | 8,286 (7.7)            | 3,986 (7.4)            | 4,300 (8.0)          |                             |
| 45–64                                | 39,834 (37.1)          | 20,733 (38.6)          | 19,101 (35.6)        |                             |
| ≥65                                  | 58,724 (54.7)          | 28,720 (53.5)          | 30,004 (55.9)        |                             |
| Body mass index Mean (kg/m²)         |                        |                        |                      |                             |
| Distribution, n (%)                  |                        |                        |                      |                             |
| <25                                  | 26.7 ± 5.1             | 26.7 ± 5.1             | 26.8 ± 5.2           | 0.134‡                     |
| 25–44                                | 63,695 (59.4)          | 31,886 (59.4)          | 31,809 (59.3)        |                             |
| Amputation during the observation period Distribution, n (%) |                        |                        |                      |                             |
| No                                   | 107,230 (99.94)        | 53,623 (99.95)         | 53,607 (99.92)       | 0.065‡                     |
| Yes                                  | 66 (0.06)              | 25 (0.05)              | 41 (0.08)            |                             |
| Breakdown of SGLT2is                 |                        |                        |                      |                             |
| Canagliflozin                        | –                      | –                      | 8,402 (15.7)         | –                           |
| Dapagliflozin                        | –                      | –                      | 10,390 (19.4)        | –                           |
| Empagliflozin                        | –                      | –                      | 16,231 (30.3)        | –                           |
| Ipragliflozin                        | –                      | –                      | 9,900 (18.5)         | –                           |
| Luseogliflozin                       | –                      | –                      | 3,600 (6.7)          | –                           |
| Tofogliflozin                        | –                      | –                      | 5,125 (9.6)          | –                           |
| Other anti-diabetic agents           |                        |                        |                      |                             |
| Insulin                              | 34,603 (32.3)          | 17,277 (32.2)          | 17,326 (32.3)        | 0.754†                     |
| Thiazolidinediones                   | 7,391 (6.9)            | 3,798 (7.1)            | 3,593 (6.7)          | 0.014†                     |
| Sulfonylureas                        | 23,563 (22.0)          | 11,731 (21.9)          | 11,832 (22.1)        | 0.461†                     |
| Glinides                             | 9,758 (9.1)            | 4,811 (9.0)            | 4,947 (9.2)          | 0.152†                     |
| α-GIs                                | 11,207 (10.4)          | 5,574 (10.4)           | 5,633 (10.5)         | 0.563†                     |
| DPP-4 inhibitors                     | 63,779 (59.4)          | 32,027 (59.7)          | 31,752 (59.2)        | 0.089†                     |
| GLP-1 receptor agonists              | 4,523 (4.2)            | 2,198 (4.1)            | 2,325 (4.3)          | 0.056‡                     |
| Antiplatelet/anticoagulant drugs      |                        |                        |                      |                             |
| Aspirin                              | 22,942 (21.4)          | 11,521 (21.5)          | 11,421 (21.3)        | 0.461†                     |
| P2Y12 inhibitors                     | 16,067 (15.0)          | 8,011 (14.9)           | 8,056 (15.0)         | 0.707†                     |
| PDE inhibitors                       | 3,945 (3.7)            | 1,991 (3.7)            | 1,954 (3.6)          | 0.559†                     |
| Prostaglandins                       | 2,242 (2.1)            | 1,153 (2.1)            | 1,089 (2.0)          | 0.179†                     |
| SHT2 inhibitors                      | 492 (0.5)              | 265 (0.5)              | 227 (0.4)            | 0.095†                     |
| Warfarin                             | 3,420 (3.2)            | 1,715 (3.2)            | 1,705 (3.2)          | 0.876‡                     |
| DOACs                                | 6,331 (5.9)            | 3,181 (5.9)            | 3,150 (5.9)          | 0.698‡                     |
| Statins                              |                        |                        |                      |                             |
| Standard statins                     | 5,138 (4.8)            | 2,635 (4.9)            | 2,503 (4.7)          | 0.061‡                     |
| Strong statins                       | 38,256 (35.7)          | 19,099 (35.6)          | 19,157 (35.7)        | 0.716‡                     |

Data are shown as the mean ± standard deviation. Body mass index is weight in kilograms divided by the square of height in meters. †χ²-test; ‡unpaired t-test. α-GI, α-glucosidase inhibitor; DOAC, direct oral anticoagulant; DPP4, dipeptidyl peptidase-4; SHT2, 5-hydroxytryptamine 2; GLP-1, glucagon-like peptide-1; PDE, phosphodiesterase; SGLT2i, sodium-glucose cotransporter 2 inhibitor.
there was no significant difference between the two; this might be associated with the increased risk of amputation in female patients using SGLT2is presumably through a failure of local circulation due to advanced atherosclerosis in the foot. Nevertheless, the very small number of patients undergoing amputation and the lack of clinical information on possible risk factors for amputation, such as smoking status and presence/absence of atherosclerotic vascular diseases, might confound the reason for the sex difference in the amputation risk, which is a limitation of the present study.

A review of the published literature showed that risk factors for lower-limb amputation associated with SGLT2is include concomitant cardiovascular diseases. Furthermore, a recent meta-analysis of randomized controlled trials in patients with type 2 diabetes undergoing lower-limb amputation divided into metformin and sodium–glucose cotransporter 2 inhibitors groups after propensity score matching

Table 2

| Characteristics | Overall (n = 66) | Metformin (n = 25) | SGLT2is (n = 41) | P-value (SGLT2is vs metformin) |
|-----------------|-----------------|-------------------|-----------------|--------------------------------|
| Sex, n (%)      |                 |                   |                 |                                |
| Male            | 39 (59.1)       | 19 (76.0)         | 20 (48.8)       | 0.054†                         |
| Female          | 27 (40.9)       | 6 (24.0)          | 21 (51.2)       |                                |
| Age             |                 |                   |                 |                                |
| Mean (years)    | 64.5 ± 13.7     | 60.1 ± 13.0       | 67.1 ± 13.5     | 0.021§                         |
| Distribution, n (%) |               |                   |                 |                                |
| <25             | 0 (0)           | 0 (0)             | 0 (0)           | 0.019‡                         |
| 25–44           | 8 (12.1)        | 5 (20.0)          | 3 (7.3)         |                                |
| 45–64           | 20 (30.3)       | 11 (44.0)         | 9 (22.0)        |                                |
| ≥65             | 38 (57.6)       | 9 (36.0)          | 29 (70.7)       |                                |
| Body mass index |                 |                   |                 |                                |
| Mean (kg/m²)    | 24.0 ± 6.0      | 22.1 ± 2.2        | 25.2 ± 7.1      | 0.023§                         |
| Distribution, n (%) |               |                   |                 |                                |
| <25             | 47 (71.2)       | 25 (100)          | 22 (53.7)       | <0.001†                        |
| ≥25             | 19 (28.8)       | 0 (0)             | 19 (46.3)       |                                |
| Breakdown of SGLT2is |              |                   |                 |                                |
| Canagliflozin   | –               | –                 | 6               | –                              |
| Dapagliflozin   | –               | –                 | 10              | –                              |
| Empagliflozin   | –               | –                 | 18              | –                              |
| Ipragliflozin   | –               | –                 | 4               | –                              |
| Luseogliflozin  | –               | –                 | 1               | –                              |
| Tofogliflozin   | –               | –                 | 2               | –                              |
| Other anti-diabetic agents |        |                   |                 |                                |
| Insulin         | 30 (45.5)       | 15 (60.0)         | 15 (36.6)       | 0.110†                         |
| Thiazolidinediones | 5 (7.6)       | 1 (4.0)           | 4 (9.8)         | 0.642‡                         |
| Sulfonylureas   | 13 (19.7)       | 6 (24.0)          | 7 (17.1)        | 0.535§                         |
| Glinides        | 4 (6.1)         | 3 (12.0)          | 1 (2.4)         | 0.148§                         |
| α-GIs           | 6 (9.1)         | 1 (4.0)           | 5 (12.2)        | 0.396§                         |
| DPP-4 inhibitors | 39 (59.1)       | 19 (76.0)         | 20 (48.8)       | 0.054†                         |
| GLP-1 receptor agonists | 4 (6.1) | 1 (4.0)           | 3 (7.3)         | 1.000‡                         |
| Antiplatelet/anticoagulant drugs |         |                   |                 |                                |
| Aspirin         | 18 (27.3)       | 8 (32.0)          | 10 (24.4)       | 0.698†                         |
| P2Y12 inhibitors | 6 (9.1)         | 3 (12.0)          | 3 (7.3)         | 0.666‡                         |
| PDE inhibitors  | 5 (7.6)         | 1 (4.0)           | 4 (9.8)         | 0.642‡                         |
| Prostaglandins  | 3 (4.5)         | 2 (8.0)           | 1 (2.4)         | 0.552‡                         |
| SHT2 inhibitors | 1 (1.5)         | 0 (0)             | 1 (2.4)         | 1.000‡                         |
| Warfarin        | 2 (3.0)         | 1 (4.0)           | 1 (2.4)         | 1.000‡                         |
| DOACs           | 8 (12.1)        | 2 (8.0)           | 6 (14.6)        | 0.700‡                         |
| Statins         |                 |                   |                 |                                |
| Standard statins | 3 (4.5)         | 2 (8.0)           | 1 (2.4)         | 0.552‡                         |
| Strong statins  | 18 (27.3)       | 4 (16.0)          | 14 (34.1)       | 0.187†                         |

Data are shown as mean ± standard deviation. Body mass index is the weight in kilograms divided by the square of height in meters. † Fisher’s exact test, ‡ unpaired t-test, α-GIs, α-glucosidase inhibitor; DOAC, direct oral anticoagulant; DPP4, dipeptidyl peptidase-4; SHT2, 5-hydroxytryptamine 2; GLP-1, glucagon-like peptide-1; PDE, phosphodiesterase; SGLT2, sodium–glucose cotransporter 2 inhibitor.
**Table 1**

| Antidiabetic drugs | No. of events | No. of events per 1,000 patient-years | Hazard ratio (95% CI) | P-value |
|--------------------|---------------|--------------------------------------|-----------------------|---------|
| Metformin          | 25            | 3.8                                  | Reference             | -       |
| SGLT2is            | 41            | 4.0                                  | 1.34 (0.80-2.24)      | 0.267   |
| Types of SGLT2is   |                |                                      |                       |         |
| Ipragliflozin      | 4             | 1.8                                  | 0.78 (0.27-2.25)      | 0.640   |
| Empagliflozin      | 18            | 5.1                                  | 1.77 (0.95-3.31)      | 0.071   |
| Canagliflozin      | 6             | 3.7                                  | 1.27 (0.52-3.14)      | 0.600   |
| Dapagliflozin      | 10            | 6.3                                  | 1.73 (0.82-3.66)      | 0.150   |
| Eptogliflozin      | 2             | 3.6                                  | 0.69 (0.16-2.95)      | 0.619   |
| Luseogliflozin     | 1             | 1.1                                  | 0.52 (0.07-3.87)      | 0.524   |

**Figure 2** | Risk of lower-limb amputation in patients with type 2 diabetes using sodium–glucose cotransporter 2 inhibitors (SGLT2is) compared with that in those using metformin. Forest plots show the hazard ratios (HRs) for lower-limb amputation in patients with type 2 diabetes. Circles represent the HR and horizontal bars extend from the lower limit to the upper limit of the 95% confidence interval (CI) of the estimated HR.

**Table 2**

| Characteristics | Metformin | SGLT2is |
|-----------------|-----------|---------|
| Age (years)     | Male      | Female  |
| <65             | 20        | 21      |
| ≥75             | 13        | 12      |
| BMI (kg/m²)     | ≥25       | <25     |
| ≥25             | 19        | 16      |
| <25             | 5         | 2       |
| DPP-4 inhibitors| 20        | 20      |
| Glinides        | 3         | 1       |
| GLP-1RA         | 3         | 2       |
| Insulin         | 15        | 15      |
| Sulfonylureas   | 6         | 7       |
| Thiazolidinediones| 1        | 4       |
| α-GIs           | 1         | 5       |
| PDE inhibitors  | 1         | 2       |
| Prostaglandin   | 2         | 1       |
| P2Y12 inhibitors| 3         | 3       |
| SHT2 inhibitors | 0         | 1       |
| DOAC            | 2         | 6       |
| Statins         | 2         | 1       |
| Strong statins  | 4         | 14      |

**Figure 3** | Comparison of the risk of lower-limb amputation between patients with type 2 diabetes using sodium–glucose cotransporter 2 inhibitors (SGLT2is) and those using metformin, according to the baseline clinical characteristics. Forest plots show the hazard ratios (HRs) for lower-limb amputation in patients with type 2 diabetes based on baseline clinical characteristics. Circles represent the HR, and horizontal bars extend from the lower limit to the upper limit of the 95% confidence interval (CI) of the estimated HR. a-GI, α-glucosidase inhibitor; BMI, body mass index; DOAC, direct oral anticoagulant; DPP4, dipeptidyl peptidase-4; SHT2, 5-hydroxytryptamine 2; GLP-1RA, glucagon-like peptide-1 receptor agonist; NA, not applicable; PDE, phosphodiesterase.
diabetes showed that the risk of amputation increased in patients using SGLT2is, which was mainly driven by the results of cardiovascular outcome trials. These findings suggest that atherosclerotic cardiovascular diseases might be closely related to SGLT2i-associated amputation. In the present study, we could not collect patient information on the history of atherosclerotic cardiovascular disease, including PAD. However, considering that the risk of amputation was significantly higher in strong statin users using SGLT2is than in those using metformin in the present study, using SGLT2is in patients with type 2 diabetes at high risk of or affected by severe atherosclerosis requiring strong statins might be a risk factor for lower-limb amputation.

Regarding obese patients with type 2 diabetes, amputation events were observed only in patients using SGLT2is, but not in those using metformin, although the reason for the null event in the metformin group remains unclear. To the best of our knowledge, few studies have shown the association between higher BMI and amputation risk in patients with type 2 diabetes using SGLT2is. Meanwhile, a recent meta-analysis showed that diabetic foot ulcer-related amputations are more likely to occur in patients with lower BMIs. Diabetic foot ulcer, especially when concomitant with local infections, is a consumptive disease. A higher BMI might reflect better nutritional status and, hence, might work in favor of coping with a severe illness. The mitigating effect of higher BMI on the risk of diabetic foot ulcer-related amputations might be what is called “the obesity paradox,” a phenomenon involving better health outcomes associated with elevated bodyweight, which could potentially explain the null event of amputation in obese patients with type 2 diabetes using metformin in the present study. However, using SGLT2is might counteract this phenomenon through the hemodynamic effect of diuresis. Alternatively, a recent meta-analysis using data from randomized controlled trials showed that greater bodyweight reduction in SGLT2i users was associated with an increased risk of amputation. The decrease in bodyweight associated with SGLT2is is reported to be greater in patients with type 2 diabetes having higher baseline BMIs; therefore, greater bodyweight reduction associated with SGLT2is might partly contribute to the increased number of amputation cases in obese patients with type 2 diabetes using SGLT2is in the present study.

The present study had several limitations that must be considered while interpreting the findings. First, limitations possibly stem from the use of diagnostic codes and prescription claims to identify patients with type 2 diabetes and ascertain amputation events. It is important to recognize that the MDV database does not represent the real conditions of all patients with diabetes, because it consists of patients treated mainly in acute care or emergency hospitals utilizing a DPC system, which mainly treat advanced-stage patients. However, given that the proportions of patients with type 2 diabetes reported in the MDV database and the Japanese Health and Nutrition Examination Survey were similar, the results of the present study might be applicable to most patients with type 2 diabetes. Second, the incidence of amputation was considerably low, possibly preventing an accurate analysis. In addition, considering that this was a retrospective cohort study, further prospective studies are necessary to evaluate the actual effect of SGLT2is on lower-limb outcomes in the future. Third, we lacked important clinical information (glycated hemoglobin level, diabetes duration, smoking status, diabetic microvascular complications, including diabetic retinopathy, and macrovascular complications, including atherosclerotic diseases), all of which could be associated with amputation risk. In particular, it was difficult to determine whether patients developed complications or not, because the disease names registered in the DPC system usually included tentative names for the purpose of the diagnostic tests, which are hardly distinguishable from the actual disease names. Thus, we could not use information on complications in the present study. Furthermore, most of the lower-limb amputations in patients with diabetes are often attributable to uncontrolled local infections and after necrosis of the extremities, which might be related to comorbidities, including diabetic foot and PAD. Such conditions might precede surgical management and ultimately lead to amputations. However, we could not obtain information regarding the presence/absence of infection, PAD or diabetic foot for the same reason stated above, possibly preventing an accurate analysis of the association between SGLT2is and lower-limb amputation.

We attempted to carry out a re-analysis by adding the information regarding hypertension and diabetic neuropathy, which are related to amputation. Patients with hypertension were defined as those prescribed with any antihypertensive drugs, including renin–angiotensin system inhibitors. Patients with diabetic neuropathy were defined as those prescribed with tricyclic antidepressants, pregabalin and/or duloxetine. As a result, even after propensity score matching, statistically significant differences were noted in most baseline clinical characteristics between the two groups, distorting an accurate analysis on the effect of hypertension or diabetic neuropathy on the risk for lower-limb amputation associated with SGLT2is, which is also a limitation of the present study.

In conclusion, SGLT2is might not be associated with an increased risk of lower-limb amputation in patients with type 2 diabetes. However, female patients with type 2 diabetes using SGLT2is might have a higher risk of amputation than those using metformin. Furthermore, the amputation risk might be higher in patients with type 2 diabetes using SGLT2is and strong statins than in those using metformin and strong statins. Thus, we would like to emphasize that clinicians should pay attention to patients’ foot conditions in routine clinical practice when they prescribe SGLT2is, particularly in female patients with type 2 diabetes and patients with type 2 diabetes with suspected advanced atherosclerosis requiring strong statins.

Conversely, we could not clarify the risk of lower-limb amputation associated with SGLT2is in patients with type 2 diabetes at high risk for cardioenal complications in the present study due to a lack of clinical data, such as duration of...
diabetes, smoking status, glycated hemoglobin levels and micro- or macrovascular complications, which is a limitation of the present study, using DPC-based clinical databases. However, considering that the use of SGLT2is is currently essential to patients with type 2 diabetes to prevent cardio renal complications, who are generally considered at high risk for micro-/macrovascular complications including PAD, clinicians should consider the risk of lower-limb amputation when using SGLT2is in patients with type 2 diabetes.

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Approval of the research protocol: The ethics board of Kitasato University admitted that the protocol for this study does not require ethics approval, because all available data are completely anonymous with no personal information, which is characteristic of DPC-based clinical databases (Control number: B19-285, dated 31 January 2020). This study was carried out in accordance with the Declaration of Helsinki, and the ethical guidelines for medical and health research involving human subjects.

Informed consent: All patient data were anonymized and contained no personal data; thus, informed consent was not required. Registry and the registration no. of the study/trial: N/A.

Animal studies: N/A.

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**SUPPORTING INFORMATION**

Additional supporting information may be found online in the Supporting Information section at the end of the article.

**Table S1** | Baseline clinical characteristics of patients with type 2 diabetes using metformin and sodium–glucose cotransporter 2 inhibitors before 1:1 propensity score matching, and the number of those who underwent lower-limb amputation during the observation period.