Sodium-glucose co-transporter-2 inhibitor use and risk of lower-extremity amputation: Evolving questions, evolving answers

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Aim: To examine whether sodium-glucose co-transporter-2 (SGLT2) inhibitors are associated with a higher risk of lower-extremity amputation than dipeptidyl-peptidase-4 (DPP-4) inhibitors and sulphonylureas.

Methods: We conducted a retrospective cohort study, using the MarketScan Commercial Claims and Encounters Database (2013-2015), to compare the incidence of lower-extremity amputation (LEA) between initiators of SGLT2 inhibitors and initiators of two second-line drugs, DPP-4 inhibitors and sulphonylureas (SUs). We estimated crude incidence rates (IRs) and adjusted hazard ratios (aHR), with 95% confidence intervals (CIs), before and after propensity-score weighting. We additionally conducted sensitivity analyses using a comparator group of all non-metformin, non-SGLT2 inhibitor glucose-lowering drugs, as previous studies used this approach.

Results: In a cohort of 328 150 individuals aged 18 to 64 years, the IR of LEA ranged from 1.5 to 2.4 per 1000 person-years. In as-treated analysis, the estimated hazard of LEA was increased among SGLT2 inhibitor initiators compared to DPP-4 inhibitor initiators (aHR 1.69, 95% CI 1.20-2.38), but not compared to SU initiators (aHR 1.02, 95% CI 0.67-1.55) or non-metformin, non-SGLT2 inhibitor initiators (aHR 1.02, 95% CI 0.54-1.93). Results were consistent in intention-to-treat analysis and across a number of sensitivity analyses.

Conclusions: Among commercially insured patients in the United States, our results suggest that initiation of SGLT2 inhibitors may increase the risk of LEA compared to initiation of DPP-4 inhibitors. Contrasting results when comparing SGLT2 inhibitor initiators to DPP-4 inhibitor and SU initiators highlight the importance of choosing appropriate comparator drugs when addressing comparative effectiveness and safety questions that can inform clinical decision-making.

KEYWORDS

cohort study, database research, lower extremity amputation, pharmacoepidemiology, SGLT2 inhibitor

1 | INTRODUCTION

Sodium-glucose co-transporter-2 (SGLT2) inhibitors reduce blood glucose in type 2 diabetes mellitus (T2DM) by inhibiting renal glucose reabsorption in the proximal tubule, thereby increasing urinary glucose excretion.^{1,2} In addition to glycaemic efficacy,^{1,3} observational

and cardiovascular outcome studies demonstrate that SGLT2 inhibitors have cardio-renal protective effects versus placebo and other glucose-lowering drugs (GLDs).⁴⁻⁷

Safety concerns, however, were raised by the Canagliflozin Cardiovascular Assessment Study (CANVAS) programme, which revealed a significant increase in the risk of overall amputation (hazard ratio

[HR] 1.97, 95% confidence interval [CI] 1.41-2.75), primarily of the toe or forefoot, among patients randomized to canagliflozin, with median follow-up of 126 weeks.⁵ This represents an important finding given that T2DM is the leading cause of non-traumatic lower-extremity amputations (LEAs).⁸ In response, the US Food and Drug Administration issued a bulletin regarding amputation risk in May 2016⁹ and a drug-labelling change in July 2017.¹⁰ Recently, several observational studies have sought to corroborate this finding in broader populations, with mixed conclusions.¹¹⁻¹⁶

To address the variation in observed risk of amputation with SGLT2 inhibitors, we implemented an active comparator new-user (ACNU) study¹⁷ to estimate and compare the risk of LEA between patients initiating SGLT2 inhibitors and patients initiating two second-line GLDs, dipeptidyl peptidase-4 (DPP-4) inhibitors and sulphonylureas (SUs), which are prescribed as alternatives to SGLT2 inhibitors. Specifically, we explored how selection of different analytical populations and comparators impacted the risk estimates of LEA with SGLT2 inhibitors, and how such choices may have contributed to differences in published findings in this domain.

2 | METHODS

2.1 | Data source

We conducted a cohort study using the Truven Analytics' IBM® Watson Health MarketScan® Commercial Claims and Encounters (CCA)E Database from 2013 to 2015. MarketScan data contain inpatient, outpatient, and pharmaceutical claims and encounters data linked with demographic and enrolment information from ~350 insurance payers in the United States.¹⁸ This database is representative of the US population with employer-based insurance and offers longitudinal follow-up of patients while working for the same employer.¹⁹ The study was determined to be exempt from full institutional review board review by the University of North Carolina at Chapel Hill. The study protocol was registered with the European Network of Centres for Pharmacoepidemiology and Pharmacovigilance (ENCePP) on October 23, 2017 (EU PAS Register Number 21368, <http://www.encepp.eu/encepp/viewResource.htm?id=25456>).

2.2 | Eligible population

The base population consisted of all MarketScan beneficiaries, aged 18 to 64 years, with at least one prescription dispensing claim for an SGLT2 inhibitor, a DPP-4 inhibitor or an SU between March 1, 2013 and September 30, 2015. To ensure new use of either comparator drug, we required patients to have ≥12 months of continuous MarketScan enrolment prior to the first eligible prescription dispensing claim, during which no prescription for any of the study drug classes could be observed (washout period).

2.3 | Exposure definition

Exposure to an SGLT2 inhibitor, a DPP-4 inhibitor or an SU drug (Table S1) was defined by at least two same-class drug prescription dispensing claims observed within a predefined window.

This “prescription window” was calculated as the first prescription's recorded days' supply, plus a 30-day grace period. By requiring a second prescription and using that prescription date as the index date, we restricted the analysis to a cohort who we were reasonably confident were taking the cohort drug. Patients were excluded if they: (a) received a prescription for either study drug prior to index date; (b) switched to the comparator drug between the first and second prescriptions; (c) received both study drugs on the index date; or (d) received an empagliflozin-linagliptin combination drug on the index date (Figure S1).

2.4 | Outcome definitions

The primary outcome of LEA was defined using a combination of International Classification of Diseases, Ninth Revision, Clinical Modification and Common Procedural Terminology procedure codes. Patients who experienced LEA between the first and second prescriptions were included in the analysis, with those events captured in the baseline amputation history. Secondary outcomes included other diabetes-related conditions and procedures (tissue and bone debridement, peripheral vascular disease, and diabetic foot ulcer) commonly considered to be direct precursors or alternatives to LEA. Codes for all outcome definitions were informed through prior literature²⁰⁻³⁰ and clinical guidance (Table S2). For secondary analyses, we excluded patients with baseline history of any outcome.

2.5 | Follow-up

The primary analysis was performed in an “as-treated” fashion (Figure S2). Follow-up began at the index date (date of second prescription) and ended when an individual experienced either an outcome of interest or a censoring event (treatment discontinuation, switch or augmentation; disenrolment; or September 30, 2015), whichever came first. Secondary outcome analyses additionally censored patients if LEA was observed prior to occurrence of a secondary outcome.

2.6 | Confounding control

We used propensity scores to control for measured confounding. We incorporated covariate groupings for patient demographics, diabetes-related comorbidities, general health comorbidities, medication use history (including prior use of other GLDs), and measures of health-care system use in the propensity-score model, measured in the year prior to cohort entry (Table 1). We also applied the adjusted Diabetes Complication Severity Index (aDCSI)^{31,32} as a proxy to control for diabetes disease severity.

2.7 | Statistical analysis

We estimated propensity scores using multivariable logistic regression to model each patient's predicted probability of receiving an SGLT2 inhibitor, conditional on his or her baseline covariates. We then weighted comparator cohorts by the propensity-score odds (PS/[1-PS]) to standardize the distribution of measured covariates to the SGLT2 inhibitor cohorts, and estimated the average treatment

TABLE 1 Baseline characteristics of the eligible initiators of sodium-glucose co-transporter-2 inhibitors compared with initiators of dipeptidyl peptidase-4 inhibitors and sulphonylureas before and after implementation of standardized mortality ratio weighting^a (365-day washout period^b)

Characteristic	SGLT2 inhibitors vs. DPP-4 inhibitors			SGLT2 inhibitors vs. SUs		
	SGLT2 inhibitor initiators (n = 49 324)	DPP-4 inhibitor initiators (n = 116 439)	Weighted DPP-4 inhibitor initiators (n = 50 189)	SGLT2 inhibitor initiators (n = 46 878)	SU initiators (n = 149 623)	Weighted SU initiators (n = 48 954)
Canagliflozin	34 700 (70.3)	—	—	32 452 (69.2)	—	—
Dapagliflozin	11 041 (22.4)	—	—	10 815 (23.1)	—	—
Empagliflozin	3583 (7.3)	—	—	3611 (7.7)	—	—
Mean (SD) age, years	52.5 (8.2)	52.8 (8.2)	52.3 (8.2)	52.3 (8.2)	51.0 (9.7)	52.3 (8.1)
Men, n (%)	26 039 (52.8)	65 941 (56.6)	25 499 (50.8)	24 468 (52.2)	78 710 (52.6)	25 019 (51.1)
Diabetes comorbidities ^b						
Adjusted DCSI, mean (std. dev.)	0.7 (1.1)	0.6 (1.1)	0.7 (1.2)	0.7 (1.1)	0.6 (1.1)	0.7 (1.1)
Nephropathy, n (%)	2274 (4.6)	4480 (3.8)	2360 (4.7)	1928 (4.1)	4714 (3.2)	2089 (4.3)
Neuropathy, n (%)	5644 (11.4)	7848 (6.7)	6006 (12.0)	4941 (10.5)	8065 (5.4)	5445 (11.1)
Retinopathy, n (%)	4910 (10.0)	7593 (6.5)	4986 (9.9)	4351 (9.3)	7153 (4.8)	4816 (9.8)
Peripheral vascular disease, n (%)	1396 (2.8)	3254 (2.8)	1459 (2.9)	1320 (2.8)	3364 (2.2)	1401 (2.9)
Prior LEA, n (%)	76 (0.2)	204 (0.2)	82 (0.2)	60 (0.1)	234 (0.2)	59 (0.1)
Debridement, n (%)	1107 (2.2)	2190 (1.9)	1180 (2.4)	993 (2.1)	2393 (1.6)	1091 (2.2)
Diabetic ulcer, n (%)	1031 (2.1)	2007 (1.7)	1105 (2.2)	950 (2.0)	2370 (1.6)	965 (2.0)
General health comorbidities ^b , n (%)						
Hypertension	35 035 (71.0)	76 079 (65.3)	35 864 (71.5)	32 882 (70.1)	90 326 (60.4)	34 576 (70.6)
Dyslipidaemia	36 678 (74.4)	76 080 (65.3)	37 260 (74.2)	34 736 (74.1)	83 601 (55.9)	36 655 (74.9)
Ischaemic heart disease	5019 (10.2)	11 658 (10.0)	5032 (10.0)	4606 (9.8)	13 082 (8.7)	4884 (10.0)
Cerebrovascular disease	1754 (3.6)	4494 (3.9)	1765 (3.5)	1645 (3.5)	5402 (3.6)	1739 (3.6)
Congestive heart failure	1075 (2.2)	3231 (2.8)	1094 (2.2)	940 (2.0)	3980 (2.7)	963 (2.0)
COPD	1292 (2.6)	3405 (2.9)	1382 (2.8)	1186 (2.5)	4307 (2.9)	1281 (2.6)
Depression	4829 (9.8)	9414 (8.1)	5173 (10.3)	4746 (10.1)	12 097 (8.1)	5139 (10.5)
History of cancer	2398 (4.9)	6054 (5.2)	2456 (4.9)	2315 (4.9)	7486 (5.0)	2392 (4.9)
Chronic kidney disease	4245 (8.6)	11 037 (9.5)	4494 (9.0)	3730 (8.0)	13 049 (8.7)	3975 (8.1)
Alcohol abuse	323 (0.7)	884 (0.8)	325 (0.6)	305 (0.7)	1461 (1.0)	294 (0.6)
Smoking	2462 (5.0)	6077 (5.2)	2551 (5.1)	2266 (4.8)	8625 (5.8)	2326 (4.8)
Prior medication use ^b , n (%)						
ACE inhibitors	22 535 (45.7)	52 698 (45.3)	22 957 (45.7)	20 266 (43.2)	62 629 (41.9)	20 984 (42.9)
ARBs	13 898 (28.2)	28 923 (24.8)	14 255 (28.4)	13 581 (29.0)	29 055 (19.4)	14 711 (30.1)
Aspirin	2779 (5.6)	4594 (3.9)	3027 (6.0)	2655 (5.7)	5029 (3.4)	2883 (5.9)
β blockers	13 763 (27.9)	30 812 (26.5)	14 054 (28.0)	12 277 (26.2)	42 967 (28.7)	12 594 (25.7)
Calcium channel blockers	9680 (19.6)	23 313 (20.0)	9911 (19.7)	8872 (18.9)	28 005 (18.7)	9260 (18.9)
Statins	31 312 (63.5)	69 582 (59.8)	31 499 (62.8)	29 479 (62.9)	75 044 (50.2)	30 989 (63.3)
Loop diuretics	3615 (7.3)	7651 (6.6)	3891 (7.8)	3181 (6.8)	8910 (6.0)	3515 (7.2)
Non-loop diuretics	16 887 (34.2)	37 665 (32.3)	17 493 (34.9)	15 707 (33.5)	44 452 (29.7)	16 615 (33.9)
Digoxin	277 (0.6)	888 (0.8)	261 (0.5)	267 (0.6)	1003 (0.7)	265 (0.5)
Cilostazol	93 (0.2)	185 (0.2)	91 (0.2)	80 (0.2)	212 (0.1)	90 (0.2)
Gabapentin	4573 (9.3)	7965 (6.8)	4866 (9.7)	4137 (8.8)	9382 (6.3)	4509 (9.2)
Pregabalin	1390 (2.8)	2177 (1.9)	1487 (3.0)	1310 (2.8)	2153 (1.4)	1579 (3.2)
Duloxetine	2117 (4.3)	3710 (3.2)	2357 (4.7)	2097 (4.5)	3643 (2.4)	2415 (4.9)
Metformin	38 231 (77.5)	98 790 (84.8)	38 552 (76.8)	36 429 (77.7)	108 068 (72.2)	38 529 (78.7)
GLP-1 receptor agonists	13 950 (28.3)	5941 (5.1)	15 168 (30.2)	10 492 (22.4)	7372 (4.9)	12 640 (25.8)
DPP-4 inhibitors	—	—	—	14 769 (31.5)	28 227 (18.9)	17 187 (35.1)
SUs	17 381 (35.2)	42 590 (36.6)	17 992 (35.8)	—	—	—
Thiazolidinediones	4787 (9.7)	14 850 (12.8)	4903 (9.8)	4278 (9.1)	9930 (6.6)	4788 (9.8)
Insulin	16 162 (32.8)	14 687 (12.6)	17 150 (34.2)	14 685 (31.3)	13 259 (8.9)	16 488 (33.7)

TABLE 1 (Continued)

Characteristic	SGLT2 inhibitors vs. DPP-4 inhibitors			SGLT2 inhibitors vs. SUs		
	SGLT2 inhibitor initiators (n = 49 324)	DPP-4 inhibitor initiators (n = 116 439)	Weighted DPP-4 inhibitor initiators (n = 50 189)	SGLT2 inhibitor initiators (n = 46 878)	SU initiators (n = 149 623)	Weighted SU initiators (n = 48 954)
Measures of healthcare utilization in year prior to index date ^b , n (%)						
Number of HbA1c tests in past year						
0	4101 (8.3)	13 706 (11.8)	3959 (7.9)	3925 (8.4)	31 605 (21.1)	4174 (8.5)
1	9564 (19.4)	31 526 (27.1)	9975 (19.9)	9295 (19.8)	44 163 (29.5)	9014 (18.4)
2	13 699 (27.8)	33 432 (28.7)	14 085 (28.1)	13 232 (28.2)	37 179 (24.8)	13 296 (27.2)
3	21 960 (44.5)	37 775 (32.4)	22 170 (44.2)	20 426 (43.6)	36 676 (24.5)	22 469 (45.9)
Number of LDL tests in past year						
0	7040 (14.3)	20 606 (17.7)	7345 (14.6)	6670 (14.2)	42 324 (28.3)	7171 (14.6)
1	16 478 (33.4)	42 377 (36.4)	16 522 (32.9)	15 532 (33.1)	54 070 (36.1)	15 061 (30.8)
2	14 185 (28.8)	31 861 (27.4)	14 720 (29.3)	13 438 (28.7)	33 046 (22.1)	14 191 (29.0)
3	11 621 (23.6)	21 595 (18.5)	11 602 (23.1)	11 238 (24.0)	20 183 (13.5)	12 531 (25.6)
Flu shot received in past year	16 077 (32.6)	34 584 (29.7)	16 374 (32.6)	15 218 (32.5)	42 827 (28.6)	15 911 (32.5)
Number of hospitalizations						
0	45 735 (92.7)	105 388 (90.5)	46 412 (92.5)	43 543 (92.9)	133 177 (89.0)	45 552 (93.1)
1	2976 (6.0)	8678 (7.5)	3122 (6.2)	2757 (5.9)	12 963 (8.7)	2839 (5.8)
2	450 (0.9)	1615 (1.4)	475 (0.9)	426 (0.9)	2392 (1.6)	399 (0.8)
≥3	163 (0.3)	758 (0.7)	180 (0.4)	152 (0.3)	1091 (0.7)	163 (0.3)
Number of days in hospital						
0	45 735 (92.7)	105 388 (90.5)	46 412 (92.5)	43 543 (92.9)	133 177 (89.0)	45 552 (93.1)
1-2	1471 (3.0)	3988 (3.4)	1542 (3.1)	1346 (2.9)	5822 (3.9)	1363 (2.8)
3-5	1268 (2.6)	3730 (3.2)	1334 (2.7)	1215 (2.6)	5657 (3.8)	1252 (2.6)
5-10	501 (1.0)	1739 (1.5)	519 (1.0)	461 (1.0)	2641 (1.8)	478 (1.0)
>10	349 (0.7)	1594 (1.4)	382 (0.8)	313 (0.7)	2326 (1.6)	309 (0.6)
Number of emergency department visits						
0	39 544 (80.2)	92 730 (79.6)	39 938 (79.6)	37 775 (80.6)	115 887 (77.5)	39 563 (80.8)
1	6751 (13.7)	16 338 (14.0)	6949 (13.8)	6280 (13.4)	22 788 (15.2)	6436 (13.1)
≥2	3029 (6.1)	7371 (6.3)	3302 (6.6)	2823 (6.0)	10 948 (7.3)	2955 (6.0)
Number of physician encounters						
0	162 (0.3)	662 (0.6)	155 (0.3)	162 (0.3)	1365 (0.9)	162 (0.3)
1-3	3152 (6.4)	12 227 (10.5)	3131 (6.2)	3019 (6.4)	20 497 (13.7)	2952 (6.0)
4-6	7665 (15.5)	22 349 (19.2)	7695 (15.3)	7333 (15.6)	28 773 (19.2)	7365 (15.0)
≥7	38 345 (77.7)	81 201 (69.7)	39 208 (78.1)	36 364 (77.6)	98 988 (66.2)	38 475 (78.6)
Number of podiatrist visits						
0	44 392 (90.0)	107 032 (91.9)	45 105 (89.9)	42 275 (90.2)	139 833 (93.5)	44 034 (89.9)
1	2309 (4.7)	4547 (3.9)	2401 (4.8)	2144 (4.6)	4904 (3.3)	2306 (4.7)
2	1078 (2.2)	2102 (1.8)	1145 (2.3)	1063 (2.3)	2210 (1.5)	1108 (2.3)
≥3	1545 (3.1)	2758 (2.4)	1538 (3.1)	1396 (3.0)	2676 (1.8)	1506 (3.1)

Abbreviations: ACE, angiotensin-converting enzyme; ARB, angiotensin II receptor blockers; COPD, chronic obstructive pulmonary disease; DCSI, diabetes complication severity index; DPP-4 inhibitor, dipeptidyl peptidase-4 inhibitors; GLP-1, glucagon-like peptide-1; HbA1c, glycated haemoglobin; LEA, lower-extremity amputation; SGLT2, sodium-glucose co-transporter-2; SU, sulphonylurea.

^a Weighted by standardizing the comparator drug initiators to the population of SGLT2 inhibitor initiators, using the propensity-score odds (PS/(1-PS)), to estimate the treatment effect in those treated.

^b All baseline characteristics measured in the 1 year (365 days) prior to date of cohort drug initiation.

effect in those treated.³³ The aim of this approach was to address the question, “Would LEA incidence have changed had all SGLT2 inhibitor initiators instead initiated a comparator drug?”. Covariate balance was assessed using the standardized mean difference.³⁴

For all outcomes, we estimated crude incidence rates (IRs) using Poisson regression, and crude and adjusted HR (aHR) and 95% CIs using weighted Cox proportional hazards models. We used weighted Kaplan-Meier methods to estimate the cumulative incidence of LEA,

as well as risk differences and 95% CIs in the 1 to 30, 31 to 90, 91 to 180, and ≥ 180 days after drug initiation in each study cohort. All analyses were performed using SAS version 9.4 (SAS Institute Inc., Cary, North Carolina).

2.8 | Sensitivity and subgroup analyses

We conducted multiple sensitivity analyses. First, we re-analysed the data using an intention-to-treat (ITT) follow-up approach for up to 1 year, whereby patients were censored only for disenrolment or at the administrative study end date. Second, similarly to previous studies,¹¹⁻¹⁴ we performed an analysis requiring only a single prescription claim to be considered exposed to a study drug. Third, to replicate comparator choices in previous studies, we conducted an analysis using any non-metformin, non-SGLT2 inhibitor GLD (SUs, thiazolidinediones, DPP-4 inhibitors, glucagon-like peptide-1 receptor agonists, and insulins) as the comparator group, excluding all patients with baseline use of these agents. Fourth, because LEA risk was specifically noted in the CANVAS programme, we repeated the analysis, restricting it to canagliflozin users.

To assess the impact of various design specifications, we repeated the analyses using 15-, 60-, and 90-day grace periods; 30-, 60-, 90-, and 180-day induction periods; 30-, 60-, 90-, and 180-day latency periods; and a 6-month washout period. We specified a range of induction and latency periods a priori to determine possible durations of time required for treatment initiation to contribute to LEA occurrence; both periods were extended to 6 months to reflect CANVAS results that reported emerging amputation risk by 6 months of follow-up. We additionally repeated the analysis with 1%, 2.5% and 5% asymmetric trimming of the propensity score distribution.³⁵

The CANVAS programme reported a particularly elevated risk of LEA among patients with an LEA history, which could have important implications for clinical decision-making. To validate this result in our cohort, we estimated LEA risk within subgroups with and without prior amputation in the year prior to cohort entry. Because baseline metformin use predicts diabetes-related cardiovascular endpoints among users of second-line GLDs,³⁶ we estimated and compared stratum-specific HR estimates among patients with and without baseline metformin use in the year prior to cohort entry. Finally, to investigate the impact of other baseline GLD use and to compare results with previous literature, we performed subgroup analyses by baseline SU use, baseline insulin use, baseline cardiovascular disease (CVD) status, and baseline chronic kidney disease (CKD) status.

3 | RESULTS

3.1 | Eligible cohort

We identified a total of 328 150 eligible users of at least one study drug in the MarketScan database between March 1, 2013 and September 30, 2015. Among study patients, we identified a total of 165 763 eligible new users in the SGLT2 inhibitor ($n = 49\,324$) versus DPP-4 inhibitor ($n = 116\,439$) comparison and 196 501 eligible new users for the SGLT2 inhibitor ($n = 46\,878$) versus SU

($n = 149\,623$) comparison. Canagliflozin, dapagliflozin and empagliflozin accounted for 69% to 70%, 22% to 23%, and 7% to 8% of SGLT2 inhibitor use, respectively (Table 1). The median (interquartile range [IQR]) follow-up (Table 2) in the as-treated analyses was slightly longer among comparator drug initiators, and ranged from 0.42 (0.21-0.76) years among SGLT2 inhibitor initiators to 0.47 (0.25-0.94) years among DPP-4 inhibitor initiators. The median (IQR) follow-up was longer in the ITT analysis, again among comparator drug initiators, ranging from 0.64 (0.31-1.05) years in SGLT2 inhibitor initiators to 1.06 (0.46-2.01) years among DPP-4 inhibitor initiators and 1.03 (0.46-1.86) years among SU initiators. Follow-up was capped at 1 year after index date to minimize this observed difference in person-time available.

Age and gender distributions were similar across cohorts (Table 1). Prior to standardized mortality ratio (SMR) weighting, we observed imbalances in several baseline comorbidities, including diabetic nephropathy, neuropathy, hypertension, dyslipidaemia, ischaemic heart disease, and baseline use of angiotensin-receptor blockers, statins and metformin; baseline imbalances were generally more pronounced in the SGLT2 inhibitor versus SU comparison. All measured covariates were balanced after SMR weighting (Figure S4).

3.2 | Primary and secondary outcomes

In the as-treated analysis (Table 2), we identified 185 LEA events in the SGLT2 inhibitor ($n = 63$) versus DPP-4 inhibitor ($n = 122$) comparison, and 234 LEA events in the SGLT2 inhibitor ($n = 50$) versus SU ($n = 184$) comparison. The majority of amputations were of the toe and metatarsal (83%), foot and ankle (7%), and lower leg (9%). Absolute LEA risks were low in all cohorts (Figure 1 and Table S3). Crude IRs were higher among SGLT2 inhibitor initiators (2.4 per 1000 person-years, 95% CI 1.8-3.0) than among DPP-4 inhibitor initiators (1.5 per 1000 person-years, 95% CI 1.2-1.8), but were similar between SGLT2 inhibitor (2.0 per 1000 person-years, 95% CI 1.5-2.6) and SU (1.8 per 1000 person-years, 95% CI 1.6-2.1) initiators.

Hazard ratio estimates were similar in the crude and adjusted analyses. After controlling for baseline confounding via SMR weighting, we observed an increased hazard of LEA among SGLT2 inhibitor initiators versus DPP-4 inhibitor initiators (aHR 1.69, 95% CI 1.20-2.38); however, no association was observed when comparing SGLT2 inhibitor with SU initiators (aHR 1.02, 95% CI 0.67-1.55). To further aid clinical decision-making on second-line therapies, we performed a post hoc analysis comparing DPP-4 inhibitor initiators with SU initiators, and observed a reduced hazard of LEA among DPP-4 inhibitor initiators (aHR 0.67, 95% CI 0.49-0.91), which is consistent with the primary analysis findings. Finally, no increased hazard of LEA was observed when comparing initiators of SGLT2 inhibitors with initiators of all non-metformin, non-SGLT2 inhibitor GLDs (aHR 1.02, 95% CI 0.54-1.93), although we observed notable differences between crude and adjusted estimates in this comparison. For secondary outcomes, we observed modest to no increases in LEA hazard (Table S4), with HR estimates ranging from 0.96 to 1.22, and all 95% CIs containing the null.

TABLE 2 Crude and adjusted hazard ratio estimates for lower-extremity amputation and with initiation of sodium-glucose co-transporter-2 inhibitors versus initiation of other second-line glucose lowering drugs, 365-day washout period, as-treated analysis

Comparison	Cohort	Number of patients	Median (IQR) follow-up time, years ^a	Total person-years	Number of events ^b	Crude incidence rate, per 1000 person-years	Crude HR (95% CI)	PS-weighted HR (95% CI)
SGLT2 inhibitors vs. DPP-4 inhibitors								
AT analysis	SGLT2 inhibitor	49 324	0.42 (0.21-0.76)	26 751	63	2.4 (1.8-3.0)	1.54 (1.13-2.10)	1.69 (1.20-2.38)
	DPP-4 inhibitor	116 439	0.47 (0.25-0.94)	82 947	122	1.5 (1.2-1.8)		
ITT analysis	SGLT2 inhibitor	49 324	0.64 (0.31-1.00)	30 362	70	2.3 (1.8-2.9)	1.45 (1.08-1.93)	1.43 (1.01-2.03)
	DPP-4 inhibitor	116 439	1.00 (0.46-1.00)	86 187	136	1.6 (1.3-1.9)		
SGLT2 inhibitors vs. SUs								
AT analysis	SGLT2 inhibitor	46 878	0.42 (0.21-0.76)	25 427	50	2.0 (1.5-2.6)	1.06 (0.77-1.45)	1.02 (0.67-1.55)
	SU	149 623	0.43 (0.22-0.91)	102 058	184	1.8 (1.6-2.1)		
ITT analysis	SGLT2 inhibitor	46 878	0.64 (0.31-1.00)	28 900	56	1.9 (1.5-2.5)	1.01 (0.75-1.36)	1.02 (0.69-1.50)
	SU	149 623	1.00 (0.46-1.00)	110 352	207	1.9 (1.6-2.1)		
SGLT2 inhibitors vs. all non-metformin, non-SGLT2 inhibitors								
AT analysis	SGLT2 inhibitor	15 687	0.35 (0.17-0.64)	7275	10	1.4 (0.7-2.6)	0.66 (0.35-1.24)	1.02 (0.54-1.93)
	Non-SGLT2 inhibitor	224 363	0.41 (0.20-0.86)	148 171	282	1.9 (1.7-2.1)		
ITT analysis	SGLT2 inhibitor	15 687	0.57 (0.28-0.97)	9152	13	1.4 (0.8-2.4)	0.66 (0.38-1.16)	1.06 (0.60-1.86)
	Non-SGLT2 inhibitor	224 363	0.99 (0.44-1.00)	162 958	340	2.1 (1.9-2.3)		

Abbreviations: AT, as-treated; CI, confidence interval; DPP-4, dipeptidyl peptidase-4; HR, hazard ratio; IQR, interquartile range; ITT, intention-to-treat; PS, propensity score; SGLT2 inhibitor, sodium-glucose cotransporter-2; SU, sulphonylurea.

^a Follow-up in ITT analyses was capped at 1 year following the index date.

^b We observed 51, 59 and 102 lower-extremity amputation events between the first and second prescription fills in the DPP-4 inhibitor, SU and non-SGLT2 inhibitor comparisons, respectively. These patients were not excluded from the primary analysis, but were considered as prior amputations for the assessment of subgroup effects for patients with and without prior amputation history. These amputations were re-introduced as analysis end-points for the sensitivity analysis where we include patients with at least 1 prescription fill.

3.3 | Sensitivity and subgroup analyses

Results were consistent across a number of sensitivity analyses (Figure 2, Tables S5 and S6). We observed a downward trend in HR estimates for ITT analysis, when requiring only one prescription to define exposure, when using longer grace periods, latency, and induction periods, and when greater trimming was performed. In the SU comparison, HR estimates increased when using 60- and 90-day induction periods, although estimates remained nonsignificant. Requiring a shorter (6-month) washout period did not substantially change HR estimates. Finally, restriction to just canagliflozin initiators yielded higher HR estimates in both the DPP-4 inhibitor and SU comparisons, but lower HR estimates versus non-metformin, non-SGLT2 inhibitor initiators (aHR 0.85, 95% CI 0.38-1.91).

Subgroup analysis results suggested possible differences in LEA risk by baseline metformin and insulin use, and history of amputation, CVD and CKD (Figure 2). In both DPP-4 inhibitor and SU comparisons, we observed higher HR estimates among patients with history of amputation, with baseline insulin use, and with history of CKD. Baseline metformin use and history of CVD were associated with higher HR estimates in the SU comparison, but lower HR estimates in

the DPP-4 inhibitor comparison. We did not observe evidence of modification by baseline SU use.

4 | DISCUSSION

In this large, ACNU cohort study among commercially insured US patients, we observed a statistically significant elevation in LEA risk among SGLT2 inhibitor initiators versus DPP-4 inhibitor initiators (aHR 1.69, 95% CI 1.20-2.38), but not versus SU initiators (aHR 1.02, 95% 0.67-1.55). Primary analysis results remained consistent across a number of sensitivity analyses. We observed modest to no increases in risk for the secondary outcomes of debridement, peripheral vascular disease, and diabetic foot ulcer. Finally, risk appeared elevated in patients with history of amputation and CKD, and baseline insulin use, although these estimates were not statistically significant.

Results from this analysis join a growing body of evidence surrounding the risk of LEA associated with use of SGLT2 inhibitors (Table 3). Recent ITT analyses conducted in the MarketScan CCAE population by Yuan et al¹¹ (aHR 0.98, 95% CI 0.68-1.41) and Ryan et al¹² (aHR 1.01, 95% CI 0.80-1.28) reported null associations of below-knee amputation risk among new users of canagliflozin versus

FIGURE 1 Standardized mortality ratio (SMR)-weighted* Kaplan-Meier curves for lower-extremity amputation after drug initiation, with 365-day washout period. *Weighted by standardizing the comparator drug initiators to the population of sodium-glucose co-transporter-2 (SGLT2) inhibitor initiators, using the propensity score odds (PS/[1-PS]), to estimate the treatment effect in the treated (ATT). DPP-4, dipeptidyl peptidase-4; SU, sulfonylureas

new users of all non-SGLT2 inhibitor GLDs.^{11,12} Using as-treated analysis, Dawwas et al¹⁶ obtained a similar result versus both DPP-4 inhibitor (aHR 0.88, 95% CI 0.65-1.15) and SU users (aHR 0.74, 95% CI 0.57-0.96), and Ryan et al¹² reported a protective effect of SGLT2 inhibitors versus all non-SGLT2 inhibitor GLDs (aHR 0.56, 95% CI 0.32-0.92).

By contrast, also using MarketScan CCAE, Chang et al¹³ (aHR 1.50, 95% CI 0.85-2.67) and Adimadhyam et al¹⁴ (aHR 1.38, 95% CI 0.83-2.31) have suggested the possibility of increased LEA risk associated with new use of SGLT2 inhibitor agents, versus new use of DPP-4 inhibitors. However, low event counts and more rigid cohort exclusions in both studies contributed to limited precision and CIs that contained the null. Finally, studies by Udel et al¹⁵ (aHR 2.01, 95% CI 0.89-4.53) and Ueda et al³⁷ (aHR 2.32, 95% CI 1.37-3.91) obtained similar results to those of the CANVAS programme in different populations, although older mean age of patients in all three studies could be an underlying contributor.

In the present study, which was planned and prespecified in 2017 prior to publication of other pharmaco-epidemiological analyses, we observed a similarly elevated risk of LEA among SGLT2 inhibitor initiators versus DPP-4 inhibitor comparators, and a similar null result versus initiators of all non-metformin, non-SGLT2 inhibitor GLDs (aHR 1.02, 95% CI 0.54-1.93). However, results from the SU and non-SGLT2 inhibitor comparisons were both substantively different from the results obtained by Chang et al,¹³ who made their comparisons against a suite of older GLDs including metformin, SUs and thiazolidinediones (aHR 2.12, 95% CI 1.19-3.77). We attribute these differences to our simultaneous inclusion of insulin (typically a marker of more severe diabetes and higher amputation risk) and exclusion of metformin (often prescribed to patients with less severe diabetes and lower amputation risk) in the non-SGLT2 inhibitor comparator group, which mirrors a commonly used comparator in previous studies.^{11,12,15}

Because the potential mechanisms behind SGLT2 inhibitor and amputation risk remain undetermined – current hypotheses relate to volume depletion and reduced tissue perfusion¹¹ – we assert that the heterogeneity of findings results primarily from differences in study design decisions. We show that inference regarding comparative LEA risk changes meaningfully when different active comparators (DPP-4 inhibitor vs. SU vs. non-metformin, non-SGLT2 inhibitor GLDs) are employed. We also highlight the important differences between patients prescribed these drugs in practice, which is underscored by the crude baseline differences observed between SGLT2 inhibitor and SU initiators (Table 1), and between SGLT2 inhibitor and non-metformin, non-SGLT2 inhibitor initiators (Table S7). Sulphonylureas, for example, while often categorized as a second-line GLD in current diabetes pharmacotherapy guidelines, may not be an ideal analytical alternative to SGLT2 inhibitors because they are less costly and may be more frequently prescribed to patients with lower socio-economic status and healthcare system access.³⁸⁻⁴⁰ Furthermore, often in restrictive, protocol-driven health systems, approaches are adopted that spare the use of more recent diabetes therapies, such as SGLT2 inhibitors, until after SU treatment has failed. This has previously been

FIGURE 2 Crude and adjusted hazard ratio estimates for lower-extremity amputation with initiation of sodium-glucose co-transporter-2 (SGLT2) inhibitors vs. initiation of other second-line glucose-lowering drugs, sensitivity and subgroup analyses. AT, as-treated; BL, baseline; DPP-4, dipeptidyl peptidase-4; Hx, history; IQR, interquartile range; ITT, intention-to-treat; SMR, standardized mortality ratio; SUs, sulphonylureas.

^aWeighted by standardizing the comparator drug initiators to the population of SGLT2i initiators, using the propensity score odds (PS/(1-PS)), to estimate the treatment effect in the treated (AT). ^bPrimary analyses performed with both induction and latency periods = 0. ^cHolding induction period constant (=0). ^dHolding latency period constant (=0). ^eSubgroup analyses adjusted for age, sex, baseline neuropathy, baseline peripheral vascular disease, baseline congestive heart failure, baseline chronic kidney disease, baseline ischaemic heart disease, and baseline metformin use

demonstrated in the Medicare population, where SUs are often used as a first-line therapy as an alternative to metformin.^{41,42} Crude imbalances between SGLT2 inhibitor and non-metformin, non-SGLT2 inhibitor initiators were even more severe (Table S7). In particular, baseline CKD imbalances suggest the possibility of channeling by contraindication. Although presence of CKD was balanced after SMR weighting, this crude imbalance suggests that “catch-all” comparator groups that combine different-line diabetes therapies are not recommended for comparative effectiveness and safety studies.

Proper selection of appropriate clinical alternatives can enhance the ability of comparative safety studies to inform clinical decision-making around prescribing of similar-line medications to treat T2DM.

Other study design decisions have also contributed to the heterogeneous results of studies conducted in the MarketScan CCAE (Table 3). First, the present study, which is the first on this topic to define diabetes drug exposure using at least two prescriptions, shows that analyses defining exposure with only one prescription claim tend to yield reduced HR estimates. Second, we showed that

TABLE 3 Study design comparison of published literature assessing the association between sodium-glucose co-transporter-2 inhibitor use and amputation risk

First author/year (database)	Exclusions: Prior insulin, CKD/ESRD, baseline CVD	Mean age, years	Index drug	Comparator drug	Exposure definition	Amputation site (includes above-knee?)	Confounding control	Analytical approach	HR (95% CI)
Neal 2017 ⁵ (CANVAS)	None	63.3	CANA	Placebo	–	Yes	Randomization	ITT	1.97 (1.41-2.75)
	None	63.3	CANA	Placebo	–	No	Randomization	ITT	1.94 (1.31-2.88)
	Amputation	63.3	CANA	Placebo	–	Yes	Randomization	ITT	1.88 (1.27-2.78)
	CVD	63.3	CANA	Placebo	–	Yes	Randomization	ITT	2.34 (1.53-3.58)
Yuan 2017 ¹¹ (MarketScan CCAE)	Amputation	53.3	SGLT2 inhibitors	All non-SGLT2 inhibitors AHA ^a	1+ Rx	No	PS matching	ITT	0.98 (0.68-1.41)
Ryan 2018 ¹² (MarketScan CCAE) ^b	None	55	CANA	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	AT	0.56 (0.32-0.92)
	None	55	CANA	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	ITT	1.01 (0.80-1.28)
	Amputation	55	CANA	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	AT	0.47 (0.25-0.83)
	Amputation	55	CANA	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	ITT	0.99 (0.77-1.27)
	Insulin	55	CANA	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	AT	1.10 (0.38-2.51)
	Insulin	55	CANA	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	ITT	1.00 (0.59-1.60)
	Insulin, amputation	55	CANA	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	AT	0.87 (0.26-2.15)
	Insulin, amputation	55	CANA	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	ITT	1.00 (0.58-1.63)
	None	55	CANA	Select non-SGLT2 inhibitors AHA ^c	1+ Rx	No	PS matching	AT	1.08 (0.65-1.75)
	None	55	CANA	Select non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	ITT	0.96 (0.77-1.21)
	Amputation	55	CANA	Select non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	AT	0.85 (0.47-1.46)
	Amputation	55	CANA	Select non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	ITT	0.93 (0.72-1.18)
	Insulin	55	CANA	Select non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	AT	1.12 (0.57-2.04)
	Insulin	55	CANA	Select non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	ITT	1.20 (0.91-1.57)
	Insulin, amputation	55	CANA	Select non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	AT	0.82 (0.36-1.68)
	Insulin, amputation	55	CANA	Select non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	ITT	1.07 (0.79-1.42)
Chang 2018 ¹³ (MarketScan CCAE)	Insulin, amputation	54.2	SGLT2 inhibitors	DPP-4 inhibitors	1+ Rx	Yes	SMR weighting	AT	1.50 (0.85-2.67)
	Insulin	54.2	SGLT2 inhibitors	DPP-4 inhibitors	1+ Rx	Yes	SMR weighting	AT	1.73 (1.01-2.98)
	Insulin, amputation	52.7	SGLT2 inhibitors	GLP-1 receptor agonists	1+ Rx	Yes	SMR weighting	AT	1.47 (0.64-3.36)
	Insulin	52.7	SGLT2 inhibitors	GLP-1 receptor agonists	1+ Rx	Yes	SMR weighting	AT	1.53 (0.70-3.34)
	Insulin, amputation	51.5	SGLT2 inhibitors	SU/metformin/TZD	1+ Rx	Yes	SMR weighting	AT	2.12 (1.19-3.77)
	Insulin	51.5	SGLT2 inhibitors	SU/metformin/TZD	1+ Rx	Yes	SMR weighting	AT	2.19 (1.27-3.77)
Adimadhyam 2018 ¹⁴ (MarketScan CCAE)	Insulin, CKD	54.7	SGLT2 inhibitors	DPP-4 inhibitors	1+ Rx	Yes	PS matching	AT	1.38 (0.83-2.31)
	Insulin, CKD, amputation	54.7	SGLT2 inhibitors	DPP-4 inhibitors	1+ Rx	Yes	PS matching	AT	1.04 (0.63-1.73)
	Insulin, CKD, CVD	54.7	SGLT2 inhibitors	DPP-4 inhibitors	1+ Rx	Yes	PS matching	AT	1.50 (0.76-2.97)
	Insulin, CKD	54.7	CANA	DPP-4 inhibitors	1+ Rx	Yes	PS matching	AT	1.15 (0.64-2.07)
	Insulin, CKD	54.7	SGLT2 inhibitors	DPP-4 inhibitors	1+ Rx	No	PS matching	AT	1.38 (0.79-2.42)
	Amputation, ESRD	54.5	SGLT2 inhibitors	DPP-4 inhibitors	1+ Rx	No	PS matching	AT	0.88 (0.65-1.15)
Dawwas 2018 ¹⁶ (MarketScan CCAE)	Amputation, ESRD	54	SGLT2 inhibitors	SU	1+ Rx	No	PS matching	AT	0.74 (0.57-0.96)

TABLE 3 (Continued)

Author/year (database)	Exclusions: Prior insulin, prior amputation, baseline CKD/ESRD, baseline CVD	Mean age, years	Index drug	Comparator drug	Exposure definition	Amputation site (includes above-knee?)	Confounding control	Analytical approach	HR (95% CI)
Present study (MarketScan CCAE)	None	52.5	SGLT2 inhibitors	DPP-4 inhibitors	2+ Rx	No	SMR weighting	AT	1.69 (1.20-2.38)
	None	52.5	SGLT2 inhibitors	DPP-4 inhibitors	2+ Rx	No	SMR weighting	ITT	1.43 (1.01-2.03)
	None	52.5	SGLT2 inhibitors	SU	2+ Rx	No	SMR weighting	AT	1.02 (0.67-1.55)
	None	52.5	SGLT2 inhibitors	SU	2+ Rx	No	SMR weighting	ITT	1.02 (0.69-1.50)
	None	52.5	SGLT2 inhibitors	All non-SGLT2 inhibitors AHA	2+ Rx	No	SMR weighting	AT	1.02 (0.54-1.93)
	None	52.5	SGLT2 inhibitors	All non-SGLT2 inhibitors AHA	2+ Rx	No	SMR weighting	ITT	1.06 (0.60-1.86)
	None	52.5	CANA	DPP-4 inhibitors	2+ Rx	No	SMR weighting	AT	1.77 (1.22-2.57)
	None	52.5	CANA	SU	2+ Rx	No	SMR weighting	AT	1.18 (0.77-1.83)
	None	52.5	CANA	All non-SGLT2 inhibitors AHA	2+ Rx	No	SMR weighting	AT	0.85 (0.38-1.91)
	None	52.5	SGLT2 inhibitors	DPP-4 inhibitors	1+ Rx	No	SMR weighting	AT	1.29 (0.95-1.77)
	None	52.5	SGLT2 inhibitors	SU	1+ Rx	No	SMR weighting	AT	0.84 (0.59-1.20)
	None	52.5	SGLT2 inhibitors	All non-SGLT2 inhibitors AHA	1+ Rx	No	SMR weighting	AT	0.86 (0.49-1.50)
	Insulin	52.5	SGLT2 inhibitors	DPP-4 inhibitors	2+ Rx	No	SMR weighting	AT	1.54 (1.06-2.23)
	Insulin	52.5	SGLT2 inhibitors	SU	2+ Rx	No	SMR weighting	AT	0.85 (0.55-1.32)
	Amputation	52.5	SGLT2 inhibitors	DPP-4 inhibitors	2+ Rx	No	SMR weighting	AT	1.54 (1.06-2.23)
	Amputation	52.5	SGLT2 inhibitors	SU	2+ Rx	No	SMR weighting	AT	0.84 (0.54-1.31)
Udell 2018 ¹⁵ (US DoD Health System)	CKD	52.5	SGLT2 inhibitors	DPP-4 inhibitors	2+ Rx	No	SMR weighting	AT	1.37 (0.92-2.05)
	CKD	52.5	SGLT2 inhibitors	SU	2+ Rx	No	SMR weighting	AT	0.69 (0.43-1.11)
	CVD	52.5	SGLT2 inhibitors	DPP-4 inhibitors	2+ Rx	No	SMR weighting	AT	1.96 (1.23-3.12)
	CVD	52.5	SGLT2 inhibitors	SU	2+ Rx	No	SMR weighting	AT	0.81 (0.46-1.43)
	Amputation	65.8	SGLT2 inhibitors	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	AT	2.01 (0.89-4.53)
	Amputation	65.8	SGLT2 inhibitors	All non-SGLT2 inhibitors AHA	1+ Rx	No	PS matching	ITT	1.99 (1.12-3.51)
	ESRD	61	SGLT2 inhibitors	GLP-1 receptor agonists	1+ Rx	No	PS matching	AT	2.32 (1.37-3.91)
	ESRD	61	SGLT2 inhibitors	GLP-1 receptor agonists	1+ Rx	No	PS matching	ITT	1.90 (1.25-2.87)
	ESRD, CVD	61	SGLT2 inhibitors	GLP-1 receptor agonists	1+ Rx	No	PS matching	AT	2.12 (0.96-4.69)
	ESRD, Amputation,	61	SGLT2 inhibitors	GLP-1 receptor agonists	1+ Rx	No	PS matching	AT	2.48 (1.14-5.40)

Abbreviations: AHA, anti-hyperglycaemic agents; AT, as-treated; CCAE, Commercial Claims and Encounters; CANA, canagliflozin; CI, confidence interval; CKD, chronic kidney disease; CVD, cardiovascular disease; DoD, Department of Defense; DPP-4, dipeptidyl peptidase-4; ESRD, end-stage renal disease; GLP-1, glucagon-like peptide-1; ITT, intention-to-treat; PS, propensity score; Rx, prescription; SGLT2, sodium-glucose co-transporter-2; SMR, standardized mortality ratio; SU, sulphonylurea; TZD, thiazolidinedione.

Select non-SGLT2 inhibitor AHAs: DPP-4 inhibitors, GLP-1 receptor agonists, other AHAs.

^a All non-SGLT2 inhibitor AHAs: DPP-4 inhibitors, GLP-1 receptor agonists, TZDs, SUs, insulin, other AHAs (acarbose, bromocriptine, miglitol, nateglinide, repaglinide).

^b Only MarketScan CCAE results shown for Ryan et al. Other results, not shown, were reported for MarketScan Medicare Supplement, MarketScan Medicaid Supplement, and Optum, and are made available by the authors at <http://data.ohdsi.org/AshHBKleAmputation/>

canagliflozin-only analyses yielded higher HR estimates versus DPP-4 inhibitor and SU initiators, but lower HR estimates versus non-metformin, non-SGLT2 inhibitor initiators. This last result is more consistent with the canagliflozin-specific HR estimates observed by Yuan et al¹¹ and Ryan et al.¹² Third, analyses using ITT follow-up typically yielded HR estimates closer to the null than those using as-treated approaches, which may have contributed to the observed differences between Yuan et al¹¹ and Ryan et al,¹² versus Chang et al,¹³ Adimadhyam et al,¹⁴ and the present study. Fourth, our subgroup analyses showed that excluding patients with prior amputation and prior CKD reduced HR estimates, whereas excluding patients with baseline insulin use increased HR estimates.

Finally, differences in propensity-score methods and confounder sets can affect the extent to which confounding by indication and diabetes severity is controlled. Kaiser et al⁴³ have previously shown that potential for residual confounding can vary substantially depending on the outcome under study. Given that studies typically define a single set of confounders, one implication is that confounding control may be more complete in studies that apply these confounders to assess amputation risk alone,^{11,13,14} versus those that use the same confounders to assess both beneficial and adverse outcomes^{5,15,16} or to assess multiple adverse outcomes with different mechanisms.³⁷ In the present study, we aimed to minimize residual confounding by including confounders specific to amputation risk, and prespecified secondary outcomes that we believe share a similar mechanism to amputation risk.

Ultimately, authors should justify study design choices to reflect the specific clinical question(s) under study, and provide transparency when communicating data for clinical and regulatory decision-making. The present study aimed to address the question, "Would LEA incidence have changed had all initiators of SGLT2 inhibitor instead initiated a similar, second-line diabetes drug?". To reflect this aim, we prespecified our primary analysis to: (a) focus on new users; (b) compare with only similar second-line therapies (DPP-4 inhibitors and SUs, rather than all non-SGLT2 inhibitor GLDs); (c) require at least two prescription dispensing claims as an indicator of adherence; (d) focus on below-knee amputation for consistency with CANVAS findings; (e) employ as-treated follow-up; and (f) perform confounding control via SMR weighting to estimate the average treatment effect in those treated. We then performed a number of sensitivity and subgroup analyses to test the robustness of our primary analysis to different study design choices and specifications, and to quantify the impacts of excluding and stratifying by prior comorbidity and GLD use histories. As analytical approaches continue to evolve around questions of LEA risk with SGLT2 inhibitor use, our understanding of SGLT2 inhibitor effectiveness and safety will continue to deepen.

The present study had several strengths. First, the ACNU study design helps to address issues related to confounding by indication by comparing patients treated with similar second-line T2DM therapies, and provides additional control for other unmeasured confounders and for various time-related biases.¹⁷ Second, propensity-score weighting adequately controlled for remaining measured baseline confounding, as shown in Table 1. We controlled for baseline use of other oral GLDs, including metformin and insulin, which were not controlled for in several previous studies.^{11,13} Third, by

restricting to patients with at least two prescriptions in close proximity, we gain confidence that patients are actually taking the study drugs. The commonly used approach of defining exposure through a single prescription claim alone can include patients who are not actually on therapy, as prescription dispensing claims do not necessarily reflect whether patients actually take the study drugs after filling the prescription. In our analysis, >30% of patients with one study drug prescription did not fill a second prescription within the allotted prescription window (Figure S1, Tables S5 and S6). These patients, who contributed 26% of follow-up time in time-to-event analyses, may represent misclassified "exposed" time. Our results indicate that requiring the second prescription, which has not previously been implemented in similar studies, reduces bias towards the null.⁴⁴

Finally, we were able to leverage the size of the MarketScan CCAE database to assess subgroup differences by a number of baseline characteristics. Multiple studies have assessed possible differences in amputation risk by CVD status, but no previous observational studies have, to our knowledge, assessed differences in HR by prior amputation history and baseline metformin use. Although differences in subgroup analyses were modest and precision of estimates was limited because of low event counts, these analyses can nevertheless offer important insight to aid physician decision-making when weighing clinical alternatives among patients with specific comorbidity and diabetes medication use histories.

The present study also had the limitations characteristic of observational studies using administrative data. First, although the MarketScan CCAE database contains data for patients aged ≥ 65 years, data for these patients are inconsistently reported in MarketScan owing to the priority of Medicare coverage among such patients. To account for these potentially missing claims data, we limited our analysis to employer-insured patients aged <65 years, for whom we are confident data are relatively complete. Thus, our findings may not be generalizable to older or unemployed populations, which may have different amputation risk. Second, although we restricted the analyses to patients aged ≥ 18 years as a proxy for patients with T2DM, patients with Type 1 diabetes mellitus (T1DM) may also be present in the study cohort. However, the treatment patterns assessed in this study are more indicative of T2DM, since oral GLDs are uncommonly prescribed in T1DM.⁴⁵ Third, the MarketScan CCAE database does not report patient vital status, which precluded modelling of death as a competing risk. However, we did not expect death to be a major competing risk given the short (<1 year) follow-up in our cohort and relatively low mortality expected among the younger, privately-insured population. Fourth, because canagliflozin has dominated SGLT2 inhibitor use in the United States thus far, we did not have sufficient power to study dapagliflozin and empagliflozin individually, and were unable to determine to what extent the class effect observed in this study extends to dapagliflozin and empagliflozin.⁴⁶ However, use of dapagliflozin and empagliflozin continues to grow, and evidence should emerge in the coming years. Finally, we acknowledge the possibility of unmeasured confounding attributable to the observational nature of this study and the lack of data on race/ethnicity and

socio-economic status. However, we were able to provide partial control through claims-based definitions of dyslipidaemia, aDCSI, smoking cessation, and chronic obstructive pulmonary disease as proxies for patient body mass index, diabetes severity and duration, and smoking status, respectively.

In conclusion, the present cohort study adds to, and contextualizes, existing evidence by demonstrating that initiation of SGLT2 inhibitors may be associated with an increased LEA risk versus initiation of DPP-4 inhibitors, but not SUs. We provide evidence that risk may be greater in patients with history of LEA, history of CKD, or baseline insulin use. Study design decisions, particularly the choice of active comparator drug, can meaningfully impact HR estimates and downstream conclusions about the comparative safety of SGLT2 inhibitor and its clinical alternatives. As data continue to accrue around SGLT2 inhibitor-prescribing in real-world patient populations, analytical approaches must continue to evolve to address questions appropriately that deepen our clinical understanding of benefits and risks with SGLT2 inhibitor use.

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CONFLICTS OF INTEREST

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(Amgen, AstraZeneca, Novo Nordisk) to the Department of Epidemiology, University of North Carolina at Chapel Hill. T.S. does not accept personal compensation of any kind from any pharmaceutical company. He owns stock in Novartis, Roche, BASF, AstraZeneca and Novo Nordisk.

Author contributions

J.Y.Y. designed the study, performed analyses, interpreted results and drafted the manuscript. T.W. contributed to study design decisions and data analyses, interpreted results and edited the manuscript. V.P. performed data extraction, and provided data analysis support and verification. E.W.G. contributed to study design decisions, interpreted results and edited the manuscript. M.J.C. helped to interpret results and edit the manuscript. J.B.B. provided clinical oversight and expertise, contributed to study design decisions, interpreted results and edited the manuscript. T.S. provided methods oversight and expertise, contributed to study design decisions, interpreted results and edited the manuscript.

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SUPPORTING INFORMATION

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