Glucagon-like peptide-1 treatment reduces the risk of diabetes-type 2 related amputations

A cohort study in Denmark

Schäfer, Zeinab; Mathisen, Andreas; Thomsen, Trine Rolighed; Rossing, Peter; Kirketerp-Møller, Klaus

Published in:
Diabetes Research and Clinical Practice

DOI (link to publication from Publisher):
10.1016/j.diabres.2023.110799

Publication date:
2023

Document Version
Publisher's PDF, also known as Version of record

diabetesresearchandclinicalpractice 202 (2023) 110799

Contents lists available at ScienceDirect

Diabetes Research and Clinical Practice

journal homepage: www.journals.elsevier.com/diabetes-research-and-clinical-practice

Glucagon-like peptide-1 treatment reduces the risk of diabetes-type 2 related amputations: A cohort study in Denmark

Zeinab Schäfer [a,*], Andreas Mathisen [a,1], Trine Rolighed Thomsen [b,c], Peter Rossing [d,e], Klaus Kirketerp-Møller [d,f]

[a] Department of Computer Science, Aarhus University, Aarhus, Denmark
[b] Department of Chemistry and Biosciences, Aalborg University, Aalborg, Denmark
[c] Bio- and Environmental Technology, Danish Technological Institute, Aarhus, Denmark
[d] Steno Diabetes Center, Copenhagen, Herlev, Denmark
[e] Department of Clinical Medicine, University of Copenhagen, Gentofte, Denmark
[f] Copenhagen Wound Healing Center, Bispebjerg University Hospital, Copenhagen, Denmark

ARTICLE INFO

Keywords:
GLP-1 treatment
Amputation risk
Type 2 diabetes

ABSTRACT

Aims: To assess the impact of Glucagon-like peptide-1 (GLP-1) agonists on the risk of lower extremity amputations in patients with type 2 diabetes mellitus (DM2).
Methods: We conducted a cohort study on 309,116 patients with DM2 using Danish National Register and Diabetes Database. We tracked the GLP-1 agonists over time along with the medication dose. Time-varying models are used to assess the risk of amputation for patients with/without GLP-1 treatment.
Results: Patients on GLP-1 treatment experience a notable reduction in the risk of amputation compared to those without the treatment with a hazard ratio (HR) of 0.5, 95% CI [0.54–0.74], indicating a statistically significant difference ($p < .005$). This risk reduction was consistent across different age groups, but notably most pronounced among middle income patients. The findings were further validated by using time-varying Cox models, which considered the patient's comorbidity history.
Conclusions: Our analysis reveals compelling evidence of a reduced risk of amputation among patients receiving GLP-1 therapy, an effect dominated by liraglutide, compared to those without the treatment, even after adjusting for various socio-economic factors. However, further investigation is required to identify and account for any other potential confounding variables that may impact the outcome.

1. Introduction

One of the debilitating side effects of an untreated or severe foot ulcer in patients with DM2 is lower extremity amputation which has an immense physiological, psychological, and economic impact on the well-being of the patient and the society [1,2]. Over the last few years, there have been studies of incretin-based therapies (IBT) such as glucagon-like peptide-1 receptor agonist (GLP-1) and sodium-glucose transport protein 2 (SGLT2) that show a reduction in the risk of amputation [3–7]. Promising results have been shown for DM2 patients with a high risk of cardiovascular diseases (CVD) [6], treatment of other diseases such as dementia, obesity, nonalcoholic fatty liver disease and nonalcoholic steatohepatitis [8–10].

Furthermore, the clinical use of GLP-1 treatment has been investigated in the context of diabetic chronic ulcers in mice, with a focus on the impact of incretin hormone GLP-1 and dipeptidyl peptidase-4 inhibitors like vildagliptin on angiogenesis and wound healing [7]. There have been promising results on ulcer improvements. Another area of recent interest is the emerging role of inflammation in type 1 and type 2 diabetes, as well as related comorbidities such as arteriosclerosis and liver dysfunction [11]. A suggested mode of action is the anti-inflammatory effect of GLP-1 through immune response modulation and is currently under investigation [12].

In this cohort study, we are interested in the risk of lower-extremity amputation in patients with DM2 with/without GLP-1 treatment. Leveraging national registers and local care data in Denmark, we

* Corresponding author.
E-mail address: zeinab.schaefer@cs.au.dk (Z. Schäfer).
[1] Currently works as a Software Engineer at Humio in Aarhus, Denmark.

https://doi.org/10.1016/j.diabres.2023.110799
Received 30 May 2022; Received in revised form 5 June 2023; Accepted 21 June 2023
Available online 28 June 2023

Table 1
Data characteristics of the patients (total number of patients is 309,116 with 7333 total cases of amputation). No complication refers to the number of patients without amputation and the percentage who have the specific medical condition or are in the specific category. The column Amputations refers to the number of cases with amputation and percentage of the cases that have the medical condition and belong to the specific category.

Characteristics& Conditions	No amputation n (%)	Amputations n (%)
Age category:		
<52 years	79,301 (26.3)	2,475 (33.8)
(52–61]	76,538 (25.4)	1,518 (20.7)
(61–70]	73,454 (24.3)	1,491 (20.3)
(70–100]	72,490 (24)	1,849 (25.2)
Gender		
Male	172,421 (57.1)	5,234 (71.4)
Female	129,362 (42.9)	2,099 (28.6)
Ethnicity		
Danish	270,372 (89.6)	6,984 (95.2)
Immigrant	30,908 (10.2)	342 (4.7)
Descendent	503 (0.2)	7 (0.1)
Cardiovascular diseases	93,648 (31)	5,957 (81.2)
Nervous system disorder	83,488 (27.7)	1,613 (22)
Peripheral Arterial disease	17,970 (6)	2,176 (29.7)
Neuropathy	21,614 (7.2)	1,956 (26.7)
Bone fracture	80,128 (26.6)	1,799 (24.3)
Retinopathy	37,618 (12.5)	2,484 (33.9)
Renal complications	26,050 (8.6)	1,579 (21.5)
Mental disorder	48,154 (16)	1,138 (15.5)
Urinary tract infection	43,717 (14.5)	1,092 (14.9)
Hyperlipidemia	59,013 (19.6)	1,646 (22.4)
Hypertension	131,332 (43.5)	3,816 (52.0)
Depression	4,024 (1.3)	72 (1.0)
Dementia	3,180 (1.1)	47 (0.6)

conducted a large-scale cohort study to assess the efficacy of medication and the influence of socio-economic factors on the target population. By utilizing comprehensive national registries and incorporating prescribed medications, diagnoses, and socio-economic features such as income level, we evaluated the impact of medication on amputation risk among 309,116 patients with type 2 diabetes.

2. Materials and methods

2.1. Data source

In Denmark, healthcare data can be accessed through three different sources: national registers, local care data, and personalized tracking data. For this study, we utilized national register data for citizens born between 1900 and 1968. All citizens in Denmark have a civil registration number (CPR: Central Person Register) that enables access to all national registered data of a citizen. These include socioeconomic features through Danish National Registration [11] (i.e., income salary, birth, family status), medical history through Danish National Patient register (LPR) (determined by ICD-10 codes[2]) and the Danish National Prescription Registry [13] (LMDB) (i.e., ACT codes).

Our cohort study included a large population of 3,500,877 individuals. Within this cohort, our focus was on patients diagnosed with DM2. After extracting data on DM2 patients and filtering out any missing or incomplete patient records, we identified a total of 309,116 patients for our analysis. Among these patients, our investigation identified 7,333 cases of amputation based on our predefined criteria. Specific codes used in this study can be found in the supplementary materials, which provide additional information on data aggregation. Only patients with an ICD registration of DE11 were included in our study. For further details, refer to Table 1.

[2] In Denmark https://medinfo.dk/sks/brows.php (based on ICD codes) is used for coding diseases.

2.2. Feature definitions

2.2.1. Diagnosis date

The diagnosis date of diabetes is estimated using data from the Danish National Patient Register (LPR) based on ICD-10 codes, Dansk Voksen Diabetes Databasen (DVDD), and the Danish National Prescription Registry (LMDB) based on ACT codes. The earliest date among the first dates registered in the LPR, DVDD and LMDB is defined as the diagnosis date for each patient.

2.2.2. End of the study (EOS)

EOS is defined as the earliest instance of amputation, whether major or minor. In cases where no amputation occurred, the EOS is set as the date of death, if available. If date of death is not recorded, the EOS is set as December 31st, 2018, which represents the end of the follow-up period for all patients.

2.2.3. Diagnoses

The diagnosis onset of the following 12 medical conditions: cardiovascular related disorders (CVD), neuropathy disorders, peripheral artery diseases, hypertension, hyperlipidemia, renal complications, urinary tract infection (UTI), retinopathy, bone-fracture, mental disorder, depression, dementia with Alzheimer, and nervous system disorder. These are considered as time-varying variables in our model.

2.2.4. GLP-1 medication and tracking

We consider the intake of GLP-1 medication as a time-varying variable which reflects the changes in the intake of the medicine over time. Therefore, as the dose of the medication and the frequency of its use is relevant for this study, we construct a table that tracks the intake of different medications in time with the corresponding dose. This is done by estimating how long medications last, given the administered dose. We use the LMDB database for tracking the medications. Notably, lags between medication intake were also considered in the construction of our GLP-1 time-varying medication. If a patient halted a treatment for a few months and resumed a year later, this was accounted for in our analysis. The medication follow-up is outlined below:

Liraglutide: Liraglutide is a commonly prescribed treatment for type 2 diabetes in our database, with 34,441 patients using it. It is taken as a daily injection, starting with 0.6 mg for the first week, then increasing to 1.2 mg, up to a maximum daily dose of 3 mg. Each pen contains 3 ml solution. However, we found that the actual dose prescribed was not always recorded correctly, so we had to make some assumptions to calculate how long a medicine would last. Liraglutide has codes for 0.6 mg, 1.2 mg, and 1.8 mg doses, which last for 30, 15, and 10 days respectively. When the prescription dose was not known, we assumed a starting dose of 0.6 mg for the first 6 days and then 1.2 mg thereafter. This means the first package would last for 18 days, and each subsequent package would last for 15 days. Other medications used in the study for tracking include Dulaglutide, Semaglutide and Exenatide.[3]

We estimate the duration and dose of medication for each patient based on administration type. This way we construct a table for every patient that contains the information on their intake of GLP-1 treatment in time, forming out a time-varying covariate. We exclude the medication Adlyxin (Lyxumia (EU)) as it's no longer prescribed in Denmark. We only consider medications prescribed within the study interval (i.e., excluding the ones prescribed before the onset of diabetes or after EOS), focusing on the time interval that we are studying the effect of GLP-1

[3] Dulaglutide: The injection of Dulaglutide is once weekly, administered at an initial dose of 0.75 mg subcutaneously. Number of patients is 1,910. Semaglutide: The maximum intake of Semaglutide is 1 mg once weekly with n=4,413. Exenatide: The recommended dose is 2 mg subcutaneously once every 7 days (weekly) and twice daily initiated at 5mg. Total number of patients for these two medications is 2,649.

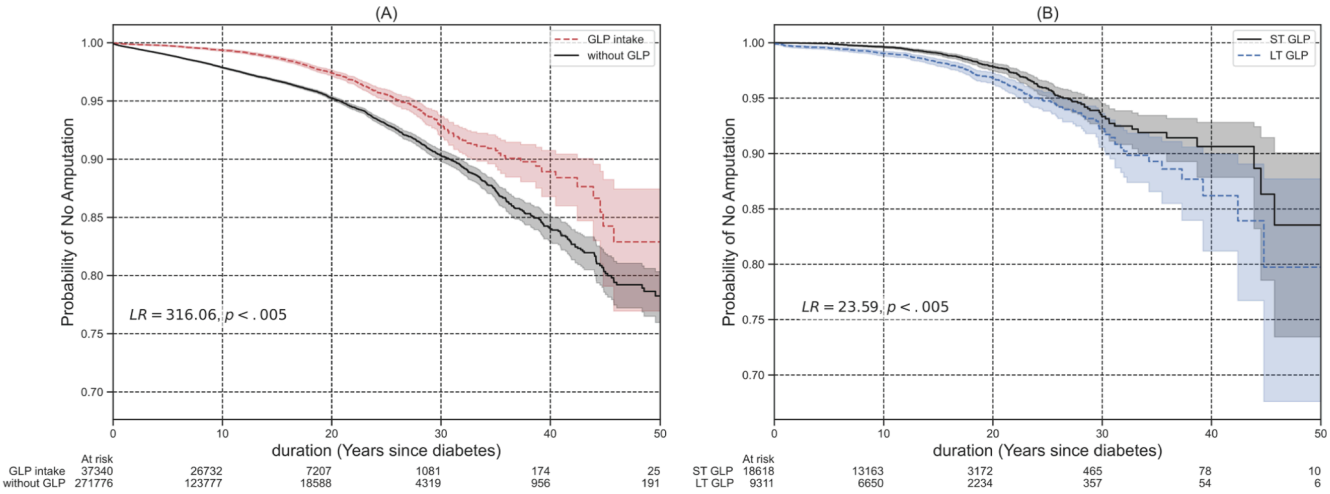

Fig. 1. (A): The cumulative probability of no amputation between patients on GLP-1 treatment versus those without the treatment over different diabetes durations. (B): Cumulative probability of amputation for patients on a short-term (ST) and long-term (LT) GLP-1 treatment. Log rank (LR) test results for both plots are stated.

treatment and if it has a protective effect against amputations. Given enough data points for each medication category, we can estimate the risk of amputation between patients on GLP-1 treatment and without the treatment.

2.2.5. Salary income & other demographic information

The Danish National Registry provides access to citizens' salary information. To calculate the family disposable income from the diabetes diagnosis until EOS, we use the average salary. In cases where complete salary information is unavailable, we estimate using either the average salary from previous years or the median net income of the total population. Patients are then categorized into three income groups: low, medium, and high.[4]

Score: The score assigned to each patient is determined by the total number of co-morbid diagnoses they have. We use the score values as a basic comparison metric between different groups, as the data does not include weighting for different disease severity.

3. Results

3.1. Amputation risk & test statistics

Fig. 1 (A) presents the amputation risk for patients on GLP-1 treatment versus those without the treatment, while (b) examines the risk among patients on short-term and long-term use of GLP-1. To assess the impact of GLP-1 intake on patients, we utilized KM estimations and categorized them into long-term and short-term treatment groups. Firstly, we excluded patients who never received any GLP-1 medication and whose study follow-up (EOS) ended before 2007, as GLP-1 prescriptions were first introduced in Denmark in that year.

Next, we calculated the total duration that patients were on GLP-1 treatment and normalized it based on the time between their diabetes onset and the study follow-up (EOS). For patients diagnosed with DM2 before 2007, we set their duration as the time from 2007 until their EOS to ensure comparability between the two groups.

Based on the normalized duration intake values, patients below the median duration (less than 27% of the time since diabetes onset or 2007) are classified as short-term users while those above the 75th percentile (more than 55% of the time since diabetes onset or 2007) are classified as long-term users. This categorization allows us to analyze the differential effects of GLP-1 treatment duration on the risk of amputation.

Figs. 2 and 3 correspond to the estimated KM curves, stratified by age and income level. Fig. 2 compares the distinct patterns between patients on GLP-1 treatment and those without the GLP-1 treatment. In Fig. 3, we specifically focus on patients receiving GLP-1 treatment (short-term and long-term). The estimated KM curves in this plot allow us to compare the effects of different treatment durations on patient outcomes within the GLP-1 treatment groups.

It is important to note that in certain plots (both in Figs. 2 and 3), the KM curves for the two groups intersect with a high standard deviation. This occurs due to the reduction in the number of amputations as the duration of GLP-1 treatment increases. With fewer patients experiencing amputations, the sample size decreases, resulting in a less precise estimate of the effect.

The average score values for patients on GLP-treatment and those without the treatment were 5.255 ± 2.13 and 4.89 ± 2.20, respectively. When considering treatment duration, short-term and long-term treatment had scores of 5.23 ± 2.13 and 5.31 ± 2.11, respectively. Before the end of the study (EOS), the average scores for groups with/without treatment were 5.22 ± 2.06 and 4.85 ± 2.11, and for short-term/long-term treatment were 5.21 ± 2.09 and 5.27 ± 2.02. These measures are used to provide insight into the overall disease burden in patients receiving GLP-1 treatment compared to those without the treatment.

3.2. Time-varying cox regression model (TVC)

To assess the risk of amputation among different groups, we conducted a further analysis using time-varying covariate (TVC) model, considering age groups and income levels. We included other medical conditions of the patients as additional time-varying covariates. Our explanatory variable, denoted as X_i, track the onset time of a particular diagnosis. Before the onset of a medical condition, we set $X_i(t) = 0$ and 1 thereafter until the EOS. We also included the intake of GLP-1 treatment as a time-varying covariate.

In our TVC model, we examined the effect of liraglutide, the most prescribed GLP-1 medication among other medications in our dataset. Fig. 4 (a, b) shows the log hazard ratios for different comorbidities. Based on the p-values, a positive log value indicates a higher risk association, while negative values suggest a protective effect. Statistical significance with $p < .001$ is denoted by a star.

Due to the limited number of patients in other medication categories and a low number of amputation cases, the standard deviations for these

[4] Low salary is considered those who earn less than half the median of general population, high salary is considered when a family earns 1.5-fold more than medium salary. Based on our data, the values are: Low salary is <= 119K DKK, High salary is >= 359K DKK, and medium salary is between low and high salaries.

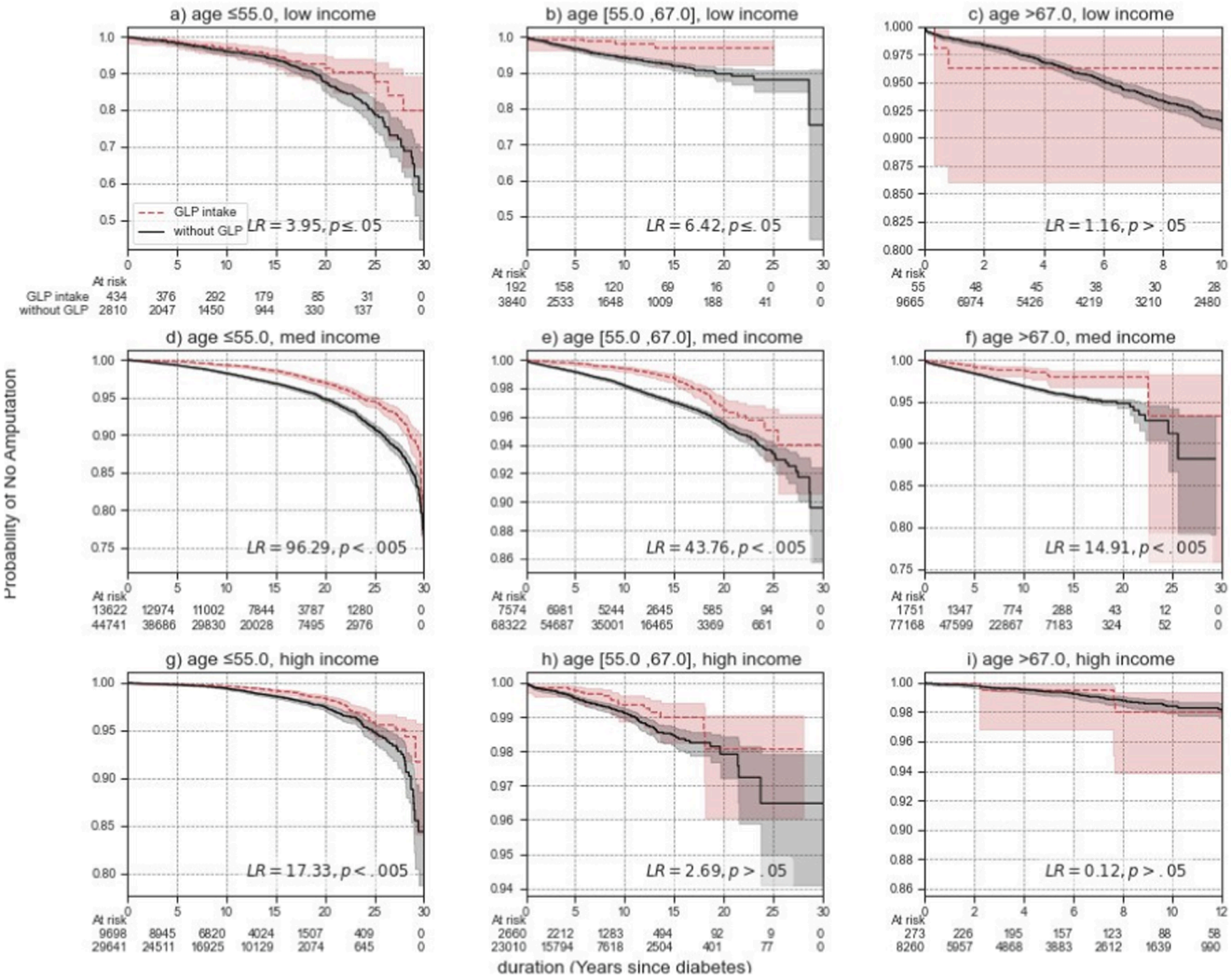

Fig. 2. The cumulative probability of no amputation for patients at different age groups and income levels between patients with/without GLP treatment.

categories were high and therefore not included in the plot for better readability. In a separate TVC model assessing the risk of amputation using different medications, we obtained the following results: Liraglutide 6 mg/ml (*coef = -0.517, CI = [-0.720,-0.315], p<.005), Semaglutide 0.25 mg (coef = -2.654,CI = [–22.08,16.77], p <.005), Semaglutide 0.5 mg (coef = 2.606, CI = [–22.364,17.152],p>.05), Exenatide 5 mg (coef = -3.444, CI = [-16.013,9.124], p>.05), Exenatide 10 mg (coef = -0.428, CI = [-1.556, 0.698], p>.05), Exenatide 2 mg (coef = 0.343, CI = [-1.043,1.729], p>.05), Semaglutide 1 mg (coef = -2.652,CI = [-35.74,30.44], p>.05), Trulicity 0.75 mg (coef = -4.363, CI = [61.79,53.06], p >.05). All models were stratified based on age and income levels, assuming distinct baseline hazards.

3.3. TVC model for major/minor amputations

We further conducted a separate analysis to examine the effect of Liraglutide treatment on major and minor amputations separately. Major amputations are related to any amputation above the ankle and minor relates to any amputation procedure below the ankle, including the foot or toe(s). Using the same methodology described in section 3.2, we calculated the likelihood of amputation for each group individually. The analysis was stratified based on age and income level. The risk of amputation for minor amputations is HR = 0.764, CI = 0.629–0.937, $p = 0.0095$ and for major amputation it is HR = 0.415, CI = 0.298–0.579, $p < 0.001$. The results suggest that Liraglutide treatment is associated with a reduced risk of both major and minor amputations.

3.4. Bias control & propensity score matching

In addition to the analyses conducted in the previous sections, we applied a propensity score matching (PSM) [14] to control for potential biases in the data. To demonstrate an example of bias, we observed that less than 2% of subjects who received GLP medication were in the low-income group, compared to 6% of subjects who did not receive GLP medication. This is the reason that we stratified the data based on age and income groups in the previous analyses. With PSM, we aimed to select subjects with similar feature distributions. Subsequently [15], we fit a logistic regression with L1 regularization on multiple features to predict the likelihood of a subject taking GLP medication. The included features were age, date of diabetes diagnosis, co-morbidities, gender, family status at the time of diabetes diagnosis, income level, and background. Based on the logits of the logistic regression, we selected up to 5 subjects without GLP medications for every subject with GLP medication. The PSM procedure resulted in an importance-weighted subset of 140,125 subjects, such that the (weighted) feature distributions of the subjects with and without GLP medications are similar. We then fit a TVC model on the weighted selected subjects similar to section 3.2, but without further stratification. The analysis revealed a HR = 0.634, CI = 0.541–0.745, $p < 0.001$, indicating the effect of Liraglutide on the likelihood of amputation.

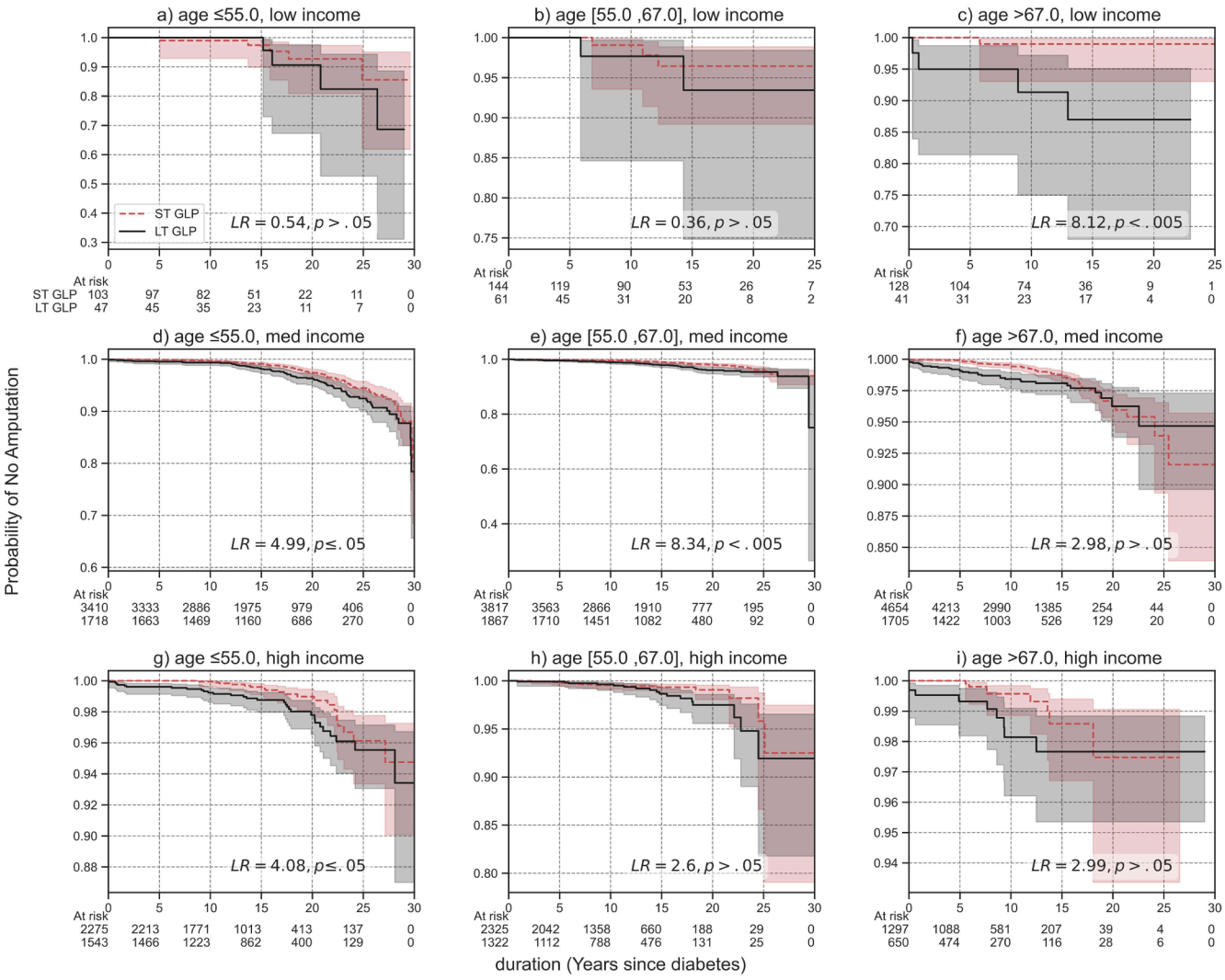

Fig. 3. The cumulative probability of no amputation between patients on short-term (ST) and long-term (LT) treatment with GLP-1 at different age and income levels.

4. Discussion

The findings of our study are consistent with some previous research in the field. Studies conducted by [6,13] have also reported similar results regarding the effectiveness of GLP-1 treatment in lowering the risk of amputation in patients with DM2. While our study did not exclusively focus on patients with type 2 diabetes at high risk of cardiovascular disease, as in [6], we did consider cardiovascular events as a confounding factor in our analysis. In a *meta*-analysis of 12 retrospective cohorts conducted by [13], the risk of lower limb amputations (LLA) was shown to be significantly higher in users of sodium-glucose cotransporter type 2 inhibitors (SGLT2is) compared to GLP-1 receptor agonists (HR 1.15, 95% CI 1.05–1.24, I2 69%). Although the primary focus of the study was not on GLP-1 agonists, the findings suggest a potential reduction in LLA among patients treated with GLP-1 (i.e., patients treated with GLP-1 have a lower risk of receiving LLA). These results contribute to the growing evidence supporting the beneficial effects of GLP-1 in reducing the risk of LLA.

We utilized the time-varying variable of GLP-1 medication to categorize patients into long-term and short-term treatment groups. Overall, our results indicated a slight reduction in amputation among patients on a short-term treatment of GLP-1(see Fig. 2). Stratifying based on age and income level; we observed similar results as shown in Fig. 3. We believe the observation may be attributed to the overall health status of the patients. It is generally believed that patients with longer-treatment durations may also have more diabetes-related complications and poorer overall health. Although the average score value between the two groups (5.23 ± 2.13 and 5.31 ± 2.11) was not significantly different, it is important to note that the score value carries equal weight for all different medical conditions, without considering the stage or severity of illness among patients in different groups.

The results are however not robust when using different definitions of long-term and short-term use of the medication. In a new setting, short-term users of GLP-1 treatment were defined as those who use the medication for less than 90 days, while long-term users are those who use are treated with GLP-1 for more than a year. The cessation of treatment after three months may be due to side effects or individuals who are unable to continue with the treatment. Although we cannot investigate the exact cause of treatment cessation, we are certain that the treatment duration was three months. With these revised definitions, our analysis indicates that patients on long-term treatment have a lower risk of amputation compared to those on short-term treatment (χ^2(df = 1, (73,4311) (401,26946)) = 4.27, p = 0.038). Stratifying the population based on age and income levels did not reveal any significant findings. Therefore, we cannot rely on these findings to determine the effectiveness of GLP-1 treatment for long-term and short-term users, as additional information regarding potential confounders and patient condition is necessary.

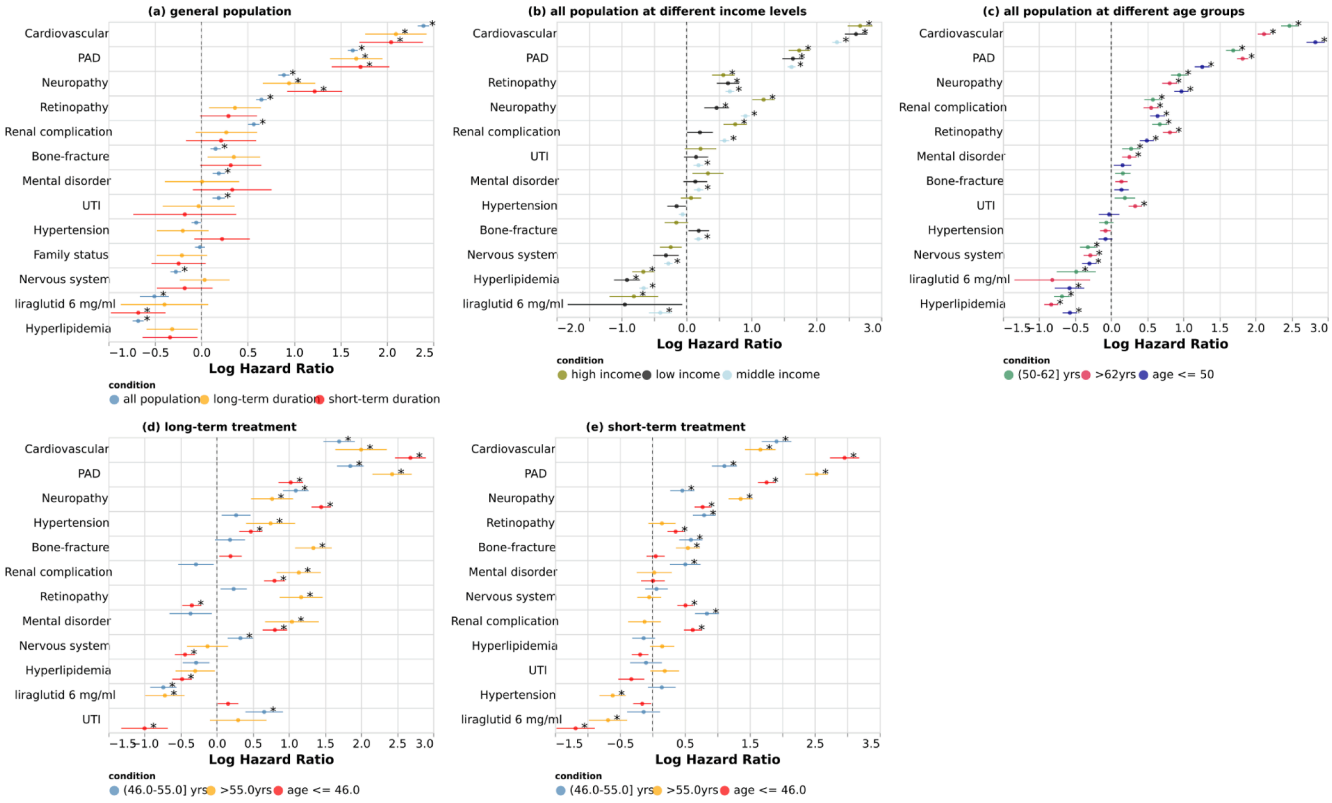

Fig. 4. The log hazard ratios of a TVC model for different medical conditions at different income levels and age groups. This is illustrated for short-term and long-term patients on GLP-1 medication and the general population.

In additional analysis of the effect of GLP-1 treatment and amputation risk, we applied a TVC model to the population of patients with major and minor amputations. The findings indicate a decreased likelihood of major amputation for patients receiving GLP-1 treatment. However, it is important to note that our data mainly covers the period from 2000 to 2018, which makes it difficult to investigate previous amputations. This is ultimately why we selected the first registration of amputation (either major or minor) as the diagnosis date for amputation, as finer modeling may introduce additional bias in the data.

Fig. 4 presents the risk of amputation in DM2 patients with different comorbidities at different income levels, age groups, and different medication duration intake. Almost across all cases (a-e), the intake of liraglutide is associated with a lower risk of amputation. These findings hold true after adjusting for income level, age groups as in plots and b.

A recurring pattern observed in all plots is the low hazard rate for condition of hyperlipidemia. This can be attributed to different confounding factors, such as other medication treatments. Patients diagnosed with hyperlipidemia often initiate statin treatment, which can have a protective effect against diabetes-related complications, consequently reducing the overall risk of amputation. In our dataset, we found that 63% of patients on GLP-1 treatment with hyperlipidemia medication were also taking statins, which may account for the lower risk observed in this group. Interaction of different medications before or during GLP-1 treatment is one of the areas of interest which we plan to assess in future.

It is important to consider that if patients on GLP-1 medication had a more severe health condition prior to starting the treatment, these results could underestimate the overall positive effect of the medication. Furthermore, due to the limited number of samples in certain groups, results carry less significance which is reflected in the standard deviation. Notably, the results for other GLP-1 medications other than liraglutide are not included in the plots due to their high standard deviation, which is attributed to the limited number of samples and a low occurrence of amputations.

4.1. Limitations

Despite numerous advantages of using national register data, there are limitations that in general are unavoidable. Examples include the annual update of data resolution for certain features, inconsistent data extraction and aggregation methods that lead to missing information, and the absence of data regarding the severity of disease. Notably, changes in the definition or computation of certain features, such as income salary can result in incomplete data. Therefore, addressing missing values is important when working with such datasets. In our study, we attempted to mitigate some of these limitations by combining different datasets and applying clear and predefined definitions to compute features (as described in Section 2).

In this study, a significant limitation related to data registry was the lack of daily medication tracking, which prevented us from measuring the level of adherence to medications over time. It is possible that individuals who adhere to their medication regimen are more likely to adopt healthier behaviors over time while those who take it for a shorter period have frail health conditions. However, when comparing score values between the two groups, the impact of medication adherence remains unclear. Different definitions of long-term and short-term intake of medication also yielded different results, which complicates the analysis further.

Additionally, the varying order in which anti-diabetic medications are prescribed to patients can contribute to differences in the overall health status of patients. It is important to examine the influence of prior medications on the effectiveness of GLP-1 treatment and its impact on the risk of amputation. Therefore, in our future studies, we intend to explore the effects of other anti-diabetic medications administered prior to GLP-1 treatment and evaluate their overall contribution to the amputation risk.

Table 2
ICD-10 codes of diagnosis used for data extraction. The codes used are maintained by the Danish Health Data Authority and can be accessed in http://medinfo.dk/sks/brows.php3.

Diagnoses	SKS code
Amputation	KNFQ (09, 19, 99), KNGQ(09, 19, 99), KNHQ(11,14,17,20,25,27,99,00,02,03,05,07)
Neuropathy disorder	DE(104,114,124,134,144), DG730 DG990, DG590, DG632, DG990
Retinopathy	DE(103,113,123,133,143), DH36
Renal disorder	DE(102,112,122,132,142), DN17
Cardiovascular disorder	DI(20,21,22,23,24,25)
Hyperlipidemia	DE(780,781,782783,784,785,786)
Hypertension	DI1
Bone fraction	DS(02,12,22,32,42,52,62,72,82,92), DT(02,08,10,12), DM(484,485,843)
Urinary tract infection	DN(109,300,309,390)
Periodic depression	DF33
Dementia	DF00
PAD	DI73
Mental disorder	DF
Nervous system disorder	DG

While the results of our study suggest a lower amputation rate in patients on GLP-1 treatment, it is important to acknowledge that there may be unadjusted confounders that could influence the findings. Therefore, the results should be interpreted with caution. To validate and strengthen these findings, further confirmation is needed through a randomized controlled trial. Currently, the STRIDE (A research study to compare a medicine called semaglutide against placebo in people with Peripheral Arterial Disease and Type 2 Diabetes) clinical trial investigates the effects of Semaglutide on walking ability compared to placebo in 800 patients with type 2 diabetes and peripheral arterial disease. Randomized studies like this one can provide more robust evidence and enhance confidence in the findings of observational cohort studies like the present study.

In the future, conducting randomized studies can provide more insights and confidence in the findings of observational cohort studies like the present study.

Declaration of Competing Interest

The authors declare the following financial interests/personal relationships which may be considered as potential competing interests: Peter Rossing has disclosed receiving consultancy and/or speaking fees (to his institution) from Astellas, AstraZeneca, Bayer, Boehringer Ingelheim, Eli Lilly, Gilead, MSD, Mundipharma, Novo Nordisk, Vifor, and Sanofi Aventis. Additionally, his institution has received research grants from AstraZeneca and Novo Nordisk.

The other authors have stated that they have no known competing financial interests or personal relationships that could have influenced the work reported in this paper.

Acknowledgements

The authors are grateful for provision of data and necessary materials from Katrine Svendsen and support of HealthD360 consortium. This study was performed within the framework of the 'HealthD360' Grand Solution project supported by Innovation Fund Denmark (no: 8056-00048A). See more here: www.healthd360.dk

Appendix A

Data sources:

Most healthcare data is scattered through different datasets. We therefore require aggregation of different datasets to obtain a meaningful database (DB). There are three main databases that are used in this study. DB1 corresponds to patient registry data (LPR) that contains all registered diagnoses since 2000 to 2018. DB2 corresponds to Dansk Voksen Diabetes Databasen (Landspatientregisteret, Lægemiddeldatabasen) that contains diabetes-related information of a group of patients before 2000 (earliest registration date is in 1973). The third DB3 corresponds to the Danish national prescription registry (LMDB, determined by ACT codes, i.e., contains the dates where anti-diabetic medications are registered and picked up by the patient) for years between 2000 and 2018.

We merge all three main databases based on CPR and set the diabetes diagnosis date to the earliest date among all entries for the patient. It is common for patients with chronic conditions like diabetes to have the same diagnosis code repeated in subsequent years. Therefore, if a patient is diagnosed in 2000 with diabetes, the same ICD code would be repeated in the following years (See Table 2).

Appendix B. Supplementary data

Supplementary data to this article can be found online at https://doi.org/10.1016/j.diabres.2023.110799.

References

[1] Davis WA, Norman PE, Bruce DG, Davis TME. Predictors, consequences and costs of diabetes-related lower extremity amputation complicating type 2 diabetes: the Fremantle Diabetes Study. Diabetologia 2006;49:2634–41. https://doi.org/10.1007/s00125-006-0431-0.

[2] Zhang Y, Lazzarini PA, McPhail SM, van Netten JJ, Armstrong DG, Pacella RE. Global Disability Burdens of Diabetes-Related Lower-Extremity Complications in 1990 and 2016. Diabetes Care 2020;43:964–74. https://doi.org/10.2337/dc19-1614.

[3] American Diabetes Association. 9. Pharmacologic Approaches to Glycemic Treatment: Standards of Medical Care in Diabetes-2019. Diabetes Care 2019;42:S90–102. https://doi.org/10.2337/dc19-S009.

[4] Marfella R, Sasso FC, Rizzo MR, Paolisso P, Barbieri M, Padovano V, et al. Dipeptidyl peptidase 4 inhibition may facilitate healing of chronic foot ulcers in patients with type 2 diabetes. Exp Diabetes Res 2012;2012:892706. https://doi.org/10.1155/2012/892706.

[5] Lovshin JA, Drucker DJ. Incretin-based therapies for type 2 diabetes mellitus. Nat Rev Endocrinol 2009;5:262–9. https://doi.org/10.1038/nrendo.2009.48.

[6] Dhatariya K, Bain SC, Buse JB, Simpson R, Tarnow L, Kaltoft MS, Stellfeld M, Tornøe K, Pratley RE; LEADER Publication Committee on behalf of the LEADER Trial Investigators. The Impact of Liraglutide on Diabetes-Related Foot Ulceration and Associated Complications in Patients With Type 2 Diabetes at High Risk for Cardiovascular Events: Results From the LEADER Trial. Diabetes Care. 2018 Oct;41(10):2229–2235. doi: 10.2337/dc18-1094. Epub 2018 Aug 2. PMID: 30072400; PMCID: PMC6150424.

[7] Nagae K, Uchi H, Morino-Koga S, Tanaka Y, Oda M, Furue M. Glucagon-like peptide-1 analogue liraglutide facilitates wound healing by activating pi3k/akt pathway in keratinocytes. Diabetes Res Clin Pract 2018;146:155–61.

[8] Sachinidis A, Nikolic D, Stoian AP, Papanas N, Tarar O, Rizvi AA, et al. Cardiovascular outcomes trials with incretin-based medications: a critical review of data available on GLP-1 receptor agonists and DPP-4 inhibitors. Metabolism 2020;111:154343. https://doi.org/10.1016/j.metabol.2020.154343.

[9] Yildirim Simsir I, Soyaltin UE, Cetinkalp S. Glucagon like peptide-1 (GLP-1) likes Alzheimer's disease. Diabetes Metab Syndr 2018 May;12(3):469–75. https://doi.org/10.1016/j.dsx.2018.03.002. Epub 2018 Mar 16. PMID: 29598932.

[10] Mantovani A, Petracca G, Beatrice G, Csermely A, Lonardo A, Targher G. Glucagon-Like Peptide-1 Receptor Agonists for Treatment of Nonalcoholic Fatty Liver Disease and Nonalcoholic Steatohepatitis: An Updated Meta-Analysis of Randomized Controlled Trials. Metabolites 2021:11. https://doi.org/10.3390/metabo11020073.

[11] Tsalamandris S, Antonopoulos AS, Oikonomou E, Papamikroulis G-A, Vogiatzi G, Papaioannou S, et al. The Role of Inflammation in Diabetes: Current Concepts and Future Perspectives. Eur Cardiol 2019;14(1):50–9.

[12] Mitchell PD, Salter BM, Oliveria JP, El-Gammal A, Tworek D, Smith SG, Sehmi R, Gauvreau GM, Butler M, O'Byrne PM. Glucagon-like peptide-1 receptor expression on human eosinophils and its regulation of eosinophil activation. Clin Exp Allergy. 2017 Mar;47(3):331–8. https://doi.org/10.1111/cea.12860. Epub 2017 Jan 28 PMID: 27928844.

[13] Scheen AJ. Lower limb amputations: protection with GLP-1 receptor agonists rather than increased risk with SGLT2 inhibitors? Diabetes Metab 2022;48(2):101325. https://doi.org/10.1016/j.diabet.2022.101325.

[14] Rosenbaum Paul R, Rubin Donald B. The central role of the propensity score in observational studies for causal effects. Biometrika 1983;70(1):41–55.

[15] Tian Yuxi, Schuemie Martijn J, Suchard Marc A. Evaluating large-scale propensity score performance through real-world and synthetic data experiments. International journal of epidemiology 2018;47(6):2005–14.