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ARTICLE

Association between glycated haemoglobin and the risk of lower extremity amputation in patients with diabetes mellitus—review and meta-analysis

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Abstract
Aims/hypothesis Diabetes increases the risk of lower extremity amputation (LEA). Although epidemiological studies report positive associations between glycaemia and LEA, the magnitude of the risk is not adequately quantified and clinical trials to date have not provided conclusive evidence about glucose lowering and LEA risk. We synthesised the available prospective epidemiological data on the association between glycaemia measured by HbA_{1c} and the risk of LEA in individuals with diabetes.
Methods We searched electronic databases and reference lists of relevant articles. We considered prospective epidemiological studies that had measured HbA_{1c} level and assessed LEA as an outcome among diabetic individuals without acute foot ulcerations or previous history of amputation. Of 2,548 citations identified, we included 14 studies comprising 94,640 participants and 1,227 LEA cases. We abstracted data using standardised forms and obtained data from investigators when required. Data included characteristics of study populations, HbA_{1c} assay methods, outcome and covariates. Study-specific relative risk estimates were pooled using random-effects model meta-analysis; heterogeneity was explored with meta-regression analyses.
Results The overall RR for LEA was 1.26 (95% CI 1.16–1.36) for each percentage point increase in HbA_{1c}. There was considerable heterogeneity across studies (I^2 76%, 67–86%; $p<0.001$), which was not accounted for by recorded study characteristics. The estimated RR was 1.44 (95% CI 1.25–1.65) for type 2 diabetes and 1.18 (95% CI 1.02–1.38) for type 1 diabetes; however, the difference was not statistically significant ($p=0.09$). We found no strong evidence for publication bias.
Conclusions/interpretation There is a substantial increase in risk of LEA associated with glycaemia in individuals with diabetes. In the absence of conclusive evidence from trials, this paper provides further epidemiological support for glucose-lowering as a strategy to reduce amputation in a population without acute foot ulceration or former amputation; it also provides disease modellers with estimates to assess the overall burden of hyperglycaemia.

Keywords Amputation · Diabetes · Disease modelling · Epidemiology · HbA_{1c} · Health economics · Hyperglycaemia · Meta-analysis · Risk factor · Systematic review

Abbreviations

LEA	Lower extremity amputation
UKPDS	UK Prospective Diabetes Study

A. I. Adler and S. Erqou contributed equally to this work.

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Introduction

Individuals with diabetes mellitus are at increased risk of macro- and microvascular complications, including, but not limited to, cardiovascular diseases, nephropathy and retinopathy. Observational studies in type 1 and type 2 diabetes have shown that these increased risks are related to the degree of glycaemic control [1, 2]. Findings from randomised trials in diabetes have confirmed that improving glycaemic control lowers the risk of microvascular complications [3–5]; however, whether it decreases the risk of cardiovascular disease is less clear [3, 6–9].

Lower extremity amputation (LEA) is a serious complication of diabetes related to both macro- and microangiopathic changes [10–12]. Diabetic individuals have a markedly increased risk of LEA when compared with non-diabetic individuals [13, 14], with potentially grave consequences; thus those with LEAs die earlier on average than those without amputations [15]. Trials of glucose lowering that have assessed its effect on the incidence of diabetic complications have generally reported LEA as part of a composite endpoint. Regardless of this, because of the low incidence of LEA, trials have not had sufficient power to detect an effect [3, 5–8, 16–18]. The PROactive and the UK Prospective Diabetes Study (UKPDS), two trials which reported on LEA as a separate endpoint, showed no difference in occurrence of LEA between groups randomised to more or less intensive glucose control (hazard ratio 1.01 [95% CI 0.58 to 1.73] and 0.70 [0.37–1.35], respectively) [3, 16, 17]. Prospective epidemiological studies, on the other hand, have suggested the presence of a graded relationship between level of glycaemia and LEA [2, 19], but individual studies did not have adequate power to estimate the magnitude of this association precisely. Although a meta-analysis of the relationship between glycaemia and cardiovascular disease from 2004 reported a positive association between glycosylated haemoglobin and peripheral vascular disease (including LEA), the estimate was based on only four studies involving fewer than 300 cases [1].

Epidemiological studies have generally used levels of fasting plasma glucose or HbA_{1c} to measure glycaemia. HbA_{1c} provides the better measure, as it reflects levels of blood glucose over several weeks, and is the main method of monitoring glycaemia in diabetes. Characterising the association between HbA_{1c} and LEA, therefore, would help understand the relationship between glycaemia and LEA and, if found to be causal, inform clinical practice by allowing clinicians to estimate the magnitude of reduction in risk that could potentially be achieved by lowering blood glucose. It would provide individuals with diabetes an estimate of the size of this association. It would also provide useful estimates to disease modellers and health economists, who analyse the cost-effectiveness of diabetes-related interventions. We report a systematic review and study-level meta-analysis of prospective epidemiological data on the association between HbA_{1c} and LEA in persons with type 1 or type 2 diabetes.

Methods

Search We systematically searched the electronic databases MEDLINE and EMBASE for studies published between January 1970 and July 2009 using key terms related to glycaemia and amputation. In the MEDLINE search, medical subject heading terms included ‘haemoglobin A, glycated’, ‘amputation’ and ‘diabetes mellitus’; key words in free text included ‘lower extremity’, ‘lower limb’, ‘amputation’, ‘HbA_{1c}’, ‘glycated haemoglobin’ and ‘glycohaemoglobin A’. We supplemented this search by scanning reference lists of relevant articles. We corresponded with investigators of included studies if the published data were not sufficient to calculate relative risks.

Study selection The search yielded 2,548 articles, which we assessed using titles, abstracts and/or full texts. The inclusion criteria were measurement of HbA_{1c} at baseline and documentation of LEA outcome during follow-up in individuals with diabetes. We took the definition of diabetes in each study as that provided in the publications. To minimise bias from reverse causation arising from the effect of existing lower extremity pathology on levels of blood glucose, we excluded cross-sectional and retrospective case–control studies, and restricted the review to studies with prospective cohort and nested case–control designs that measured HbA_{1c} at least an average of 6 months before occurrence of LEA. We excluded studies conducted in patients with acute foot ulcers, previous amputation or end-stage renal disease. When a study published more than one paper, we included the publication with the longest follow-up or largest sample size. In order to maximise the available information, we retained three studies [20–22] that combined endpoints such as peripheral vascular disease with amputations, assessing the effect of this inclusion through subgroup analysis. We selected 17 studies for inclusion and corresponded with the authors of five [21, 23–26], of whom two [21, 23] provided data, enabling us to calculate relative risks for the 14 studies in this review (Fig. 1).

Data abstraction We abstracted data using standardised forms and obtained information on study design, study year, length of follow-up, average age of participants, percentage of men, whether type 1 or type 2 diabetes, duration of diabetes, method of measuring HbA_{1c}, mean

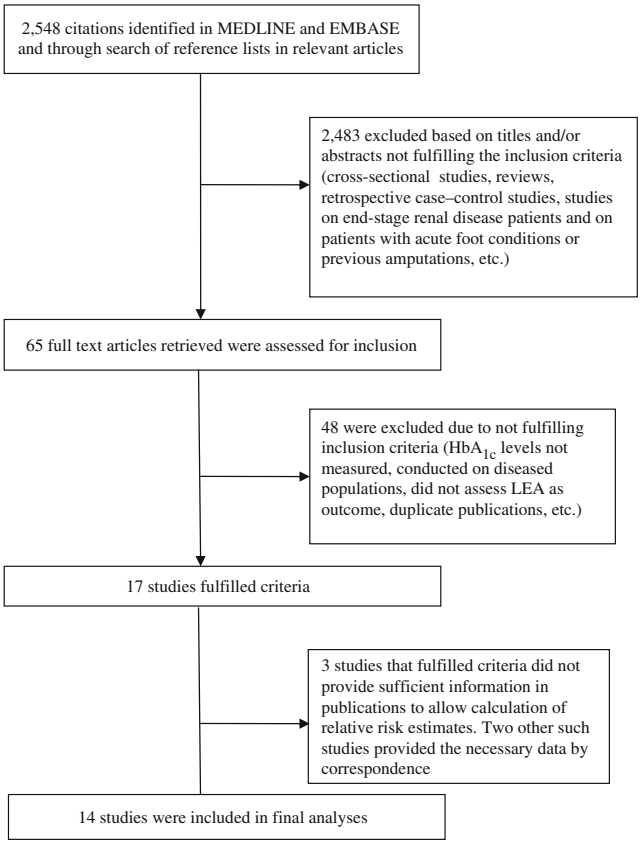

Fig. 1 Study flow diagram

level of HbA_{1c} in controls, values of relative risk for the association between HbA_{1c} and LEA (along with the unit of comparison, e.g. top vs bottom fifths), and any covariates included in regression models (e.g. age and sex). When studies reported more than one HbA_{1c} measurement, we chose the earliest to ensure the longest exposure. We rated a study's quality using the Newcastle–Ottawa quality assessment scale, a system for rating the quality of non-randomised studies in meta-analyses based on three perspectives: selection, comparability of groups and ascertainment of exposure and/or outcome [27].

Data analyses Because studies used different comparisons for HbA_{1c} (e.g. top vs bottom fourth, increase of 1 SD), we converted the risk estimates into common metrics before combining them in meta-analysis. We present risk ratios for each one percentage point increase and for the top third vs the bottom third of HbA_{1c}. We converted the risk ratios by assuming an approximately normal distribution of HbA_{1c} and a log-linear relationship between HbA_{1c} and the risk of LEA [28]. To obtain the conversion factors required to transform the relative risks, we determined the distance in SDs between the means of the quantiles using the standard normal curve. Accordingly, the log risk ratio of LEA among individuals in the top third vs the bottom third of HbA_{1c} distribution was calculated as 2.18 times the log risk ratio for a 1 SD difference in HbA_{1c} values or 2.18/2.54 times the log risk ratio for the comparison of the top and bottom fourths etc. We calculated the log ratio of the risk of a one percentage point increase in HbA_{1c} levels similarly. When we could not obtain the SD from a published report, we assumed a SD of 1.8% obtained from pooled studies. To obtain a summary estimate, we combined the estimates of relative risk using a random-effects model meta-analysis [29]. We performed a fixed-effect meta-analysis for comparison.

We assessed heterogeneity between studies using Q and I^2 statistics. The I^2 statistic estimates the percentage of total variation across studies due to a true difference rather than chance [30]. In general, I^2 values greater than 60–70% indicate the presence of substantial heterogeneity. We explored sources of heterogeneity using meta-regression and subgroup analyses. Subgroups included duration of follow-up, diabetes type, level of adjustment for confounders, type of LEA outcome and average HbA_{1c} level, as well as study design, year and quality. Data were insufficient to assess differences between subgroups defined by neuropathy, adiposity or smoking status. We assessed the presence of publication bias by comparing the combined risk estimates from larger- vs smaller-sized studies, and by using funnel plots and the Egger test of bias [31]. Descriptive statistics (e.g. age of participants, duration of follow-up etc.) are presented as ranges or weighted averages for studies that published these details. All analyses were performed using Stata release 9 (Stata, College Station, TX, USA). Statistical tests were two-sided and used a significance level of $p<0.05$.

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Results

Description of studies Fourteen prospective studies [2, 10, 12, 20–23, 32–38] involving 94,640 participants and 1,227 LEA cases were included. Details of study characteristics are provided (Table 1). The studies were North American and European, with the exception of two, which were conducted in Australia [38] and Jordan [37]. Three studies [10, 22, 37] had a nested case–control design, the rest had a cohort design. The proportion of men in the studies ranged between 33% and 98% (weighted average 85%). The average age of the participants by study ranged between 26 and 69 years (weighted average 49 years). Five studies were conducted in patients with type 1 diabetes, three in those with type 2 and the rest in mixed or unclassified diabetic populations. The average duration of diabetes in the studies ranged from 4 to 21 years. The participants were

Table 1 Characteristics of 14 prospective epidemiological studies included in the review of the association between HbA_{1c} levels and the risk of lower extremity amputation

First author [reference]	Study name	Country	Baseline year	Follow-up (years)	Participants (*n*)	Sex (% men)	Age, years (SD)	Diabetes type	Diabetes duration, years (SD)	Outcome assessed	Cases (*n*)	HbA_{1c} assay	Average HbA_{1c} in controls (%)	HbA_{1c} SD in controls (%)	Quality score[a]
Tseng [32]	VHS	USA	1998–2000	–	68,150	98	–	–	–	LEA	510	–	–	–	7
Moss [33]	WESDR	USA	1980–1982	14	1,775	47	46 (12)	Both[b]	12 (8.5)	LEA	118	Microcolumn	10.2	2.05	6
Resnick [34]	SHS	USA	1989–1992	8	1,974	36	57	–	–	LEA	87	–	8.4	–	9
Lepore [23]		Italy	2002–2003	1	172	33	69 (11)	Type 2	21 (10)	LEA	72	–	7.9	1.3	5
Mühlhauser [35]		Germany	1978–1994	10	3,570	48	27.5 (9.5)	Type 1	10.5 (9.5)	LEA	71	Microcolumn, HPLC	8.2	1.9	7
Olson [20]	PEDCS	USA	1986–1988	10	586	51	26 (8)	Type 1	18 (7)	LEA, PVD	70	Microcolumn, HPLC	10.3	1.8	7
Lehto [36]		Finland	1982	7	1,044	56	58 (0.2)	Type 2	8 (0.1)	LEA	58	Affinity chromatography	9.8	0.7	8
Jbour [37]		Jordan	2001	1	1,142	52	56 (10)	Type 2	9 (7)	LEA	53	HPLC	7.4	1.4	6
Davis [38]	FDS	Australia	1993–1996	9	1,294	48	64 (11)	Type 2	4	LEA	44	–	7.4	–	9
Stratton [2]	UKPDS	UK	1987	10	3,642	60	53 (8)	Type 2	7.5–12.5	LEA, PVD death	41	HPLC	7.1	1.8	8
Adler [12]	SDFS	USA	1988	3	776	98	65	Both[c]	9	LEA	30	–	–	–	6
Watts [10]		USA	–	–	137	51	67 (10.5)	–	–	LEA	27	–	11.5	2.6	9
Roy [21]		USA	–	6	483	40	28 (11)	Type 1	10 (9)	LEA, PVD	26	HPLC	13.5	4.3	8
Coppini [22]		UK	1982–1985	12	9,895	54	50	Both[d]	6	LEA, PVD	20	–	11.2	2.7	6

[a] For quality assessment, the Newcastle–Ottawa quality assessment scale was used, maximum score 9

[b] Type 1, 49%, type 2, 51%; [c] type 1, 7%, type 2, 93%; [d] type 1, 37%, type 2, 63%

FDS, Fremantle Diabetes Study; PEDCS, Pittsburgh Epidemiology of Diabetes Complications Study; PVD, peripheral vascular disease; SDFS, Seattle Diabetic Foot Study; SHS, Strong Heart Study; VHS, Large Veteran Health Survey & Diabetes Epidemiology Cohort; WESDR, Wisconsin Epidemiologic Study of Diabetic Retinopathy

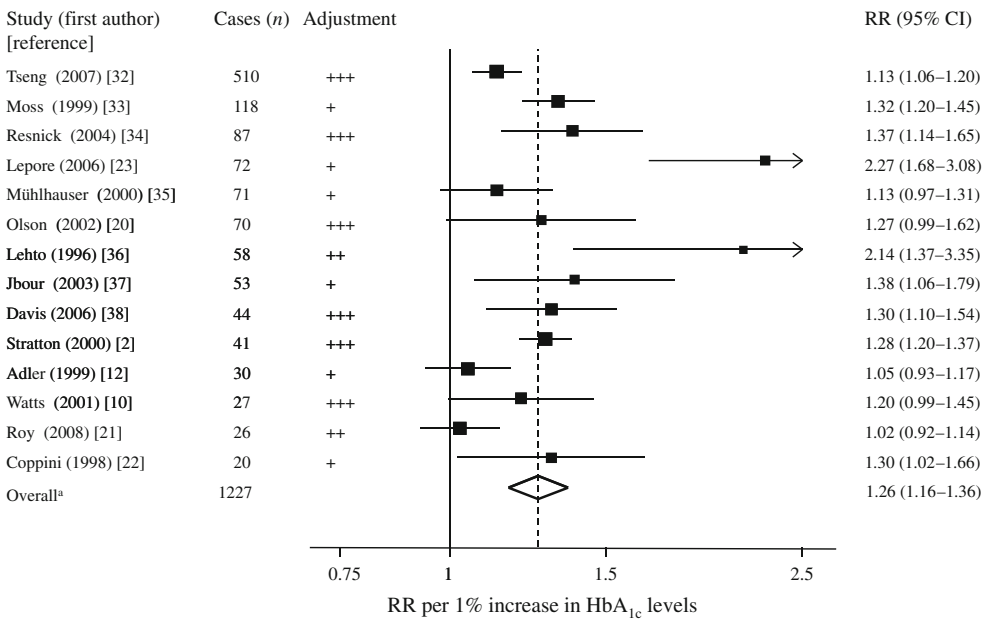

Fig. 2 Plot of RRs of amputation associated with a 1% increase in HbA_{1c} among diabetic individuals in 14 studies. [a]Overall estimate was calculated using random-effects model meta-analysis. $p<0.001$ for heterogeneity, I^2 75% (95% CI 63–84%). +, unadjusted estimates; ++, age- and sex-adjusted estimates only; +++, estimates adjusted for additional risk factors

followed for an average of 1 to 14 years. Of the reported HbA_{1c} assay methods, high performance liquid chromatography and micro-column techniques were the commonest. The average HbA_{1c} level across studies was 9.5% in controls and 11.6% in cases.

Association of HbA_{1c} with LEA Based on a random-effects model meta-analysis, the combined risk ratio for LEA associated with a one percentage point increase in HbA_{1c} was 1.26 (95% CI 1.16–1.36) (Fig. 2), with significant heterogeneity observed across studies ($p<0.001$). The corresponding estimate using a fixed-effect model meta-analysis was 1.20 (95% CI 1.17–1.24). Among studies that reported the type of diabetic population, the estimates appeared stronger for type 2 diabetes (RR 1.44, 1.25–1.65) than for type 1 diabetes (RR 1.18, 1.02–1.38) (Fig. 3), but the difference was not statistically significant ($p=0.09$). Comparing individuals in the top vs bottom third of the baseline distribution of HbA_{1c} gave a pooled risk ratio for LEA of 2.51 (95% CI 1.93–3.25) (Fig. 4).

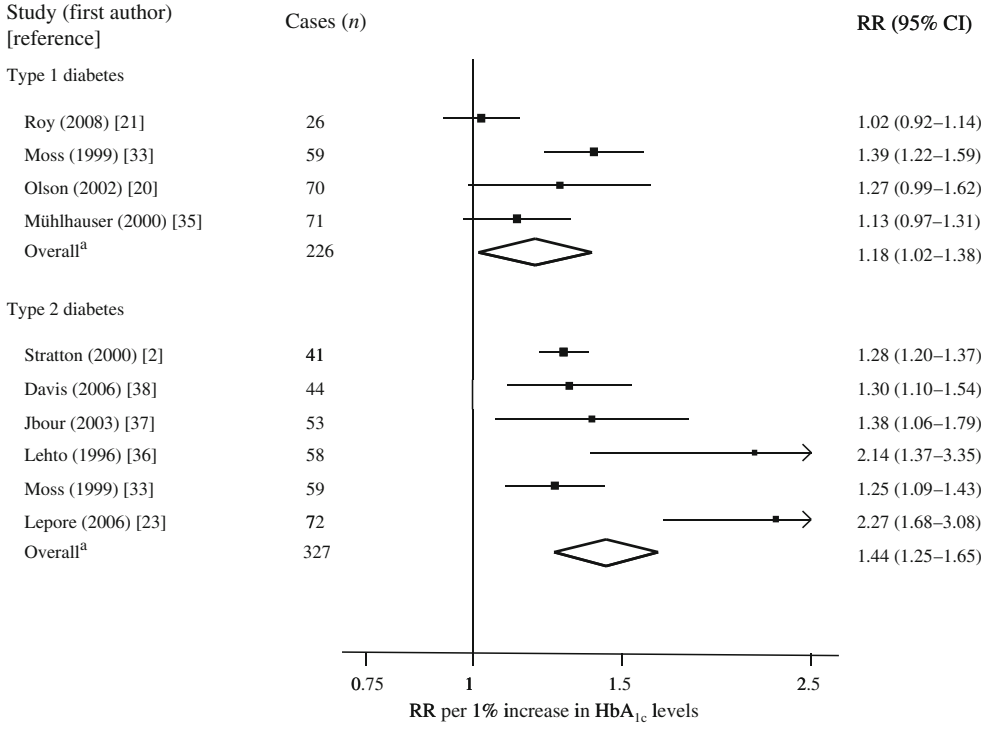

Fig. 3 Plot of RRs of amputation associated with a 1% increase in HbA_{1c} among diabetic individuals by type of diabetes. [a] Risk ratios were pooled within subgroups of diabetes type using random-effects model meta-analysis

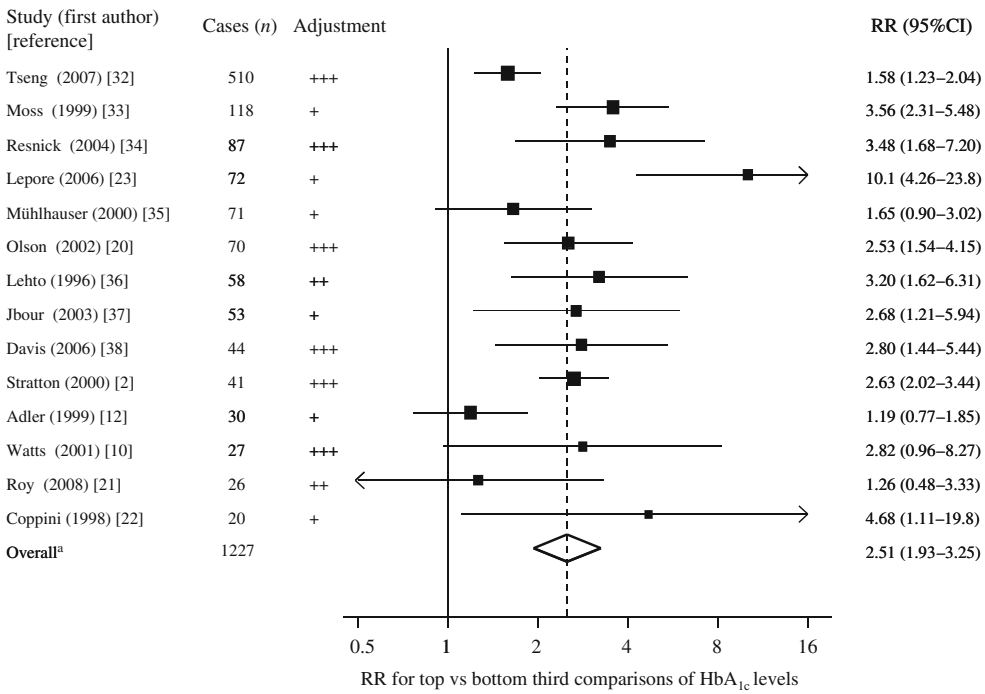

Fig. 4 Plot of RRs of amputation comparing diabetic individuals in top vs bottom thirds of baseline HbA_{1c} distribution in 14 studies. [a]Overall estimate was calculated using random-effects model meta-analysis. +, unadjusted estimates; ++, age- and sex-adjusted estimates only; +++, estimates adjusted for additional risk factors

Exploration of heterogeneity Most of the variation observed was attributable to true heterogeneity rather than sampling error as indicated by an I^2 value of 76% (95% CI 67–86%). This heterogeneity was not explained by the characteristics available for subgroup analysis (Fig. 5). We also evaluated the role of each of absolute HbA_{1c} level, duration of follow-up and duration of diabetes in meta-regression models as continuous variables, but found no significant differences. Sensitivity analyses excluding the single study that did not report a SD or the three nested case–control studies gave results consistent with the main analyses.

To assess the effect of potential publication bias, we plotted each study's risk ratio against its standard error, which suggested that smaller studies gave more extreme results (Electronic supplementary material [ESM] Fig. 1). However, the Egger test did not reach statistical significance ($p=0.085$). Comparison of the pooled estimate from larger studies (greater than the median number of cases) with that of smaller studies yielded no significant difference (Fig. 5).

Discussion

The current data provide further support for a positive association between the risk of LEA and level of HbA_{1c}. Each one percentage point increase in HbA_{1c} was associated with a 26% increase in risk of LEA, but the increase may have been as large as 36%. The risk was not significantly different for individuals with type 1 or type 2 diabetes, although the point estimate was larger for type 2 diabetes. The relationship did not vary by the study quality or HbA_{1c} concentrations, being similar in patients with moderately or extremely elevated HbA_{1c} levels. Although the average HbA_{1c} level in the present meta-analysis exceeds that of many modern diabetic populations, this suggests that the current findings may be equally true for individuals with different levels of glycaemic control. However, the analyses may not have detected a small difference or a non-linear association between HbA_{1c} and LEA. At 26%, the magnitude of the risk increase was intermediate between that reported for cardiovascular complications (18%, 95% CI 10–26%) [1] and for microvascular complications (37%, 95% CI 33–41%) [2], while acknowledging overlapping confidence intervals and that different studies contributed to the estimates. This may reflect the notion that microvascular and macrovascular disease underlie the pathogenesis of LEA via microcirculatory defects, neuropathy and arterial disease. While many consider LEA to be a late-stage complication of diabetes, we found no differences in risk by duration of disease. We did find significant heterogeneity, which could influence the generalisability of results, but we did not find strong evidence for publication bias.

The relevance of these findings is increased by the fact that the scientific evidence for glycaemia and risk of LEA to date is not sufficient to translate into clinical practice. Data from observational studies in diabetes have documented positive associations of glycaemia with microvascular and macrovascular complications, suggesting a

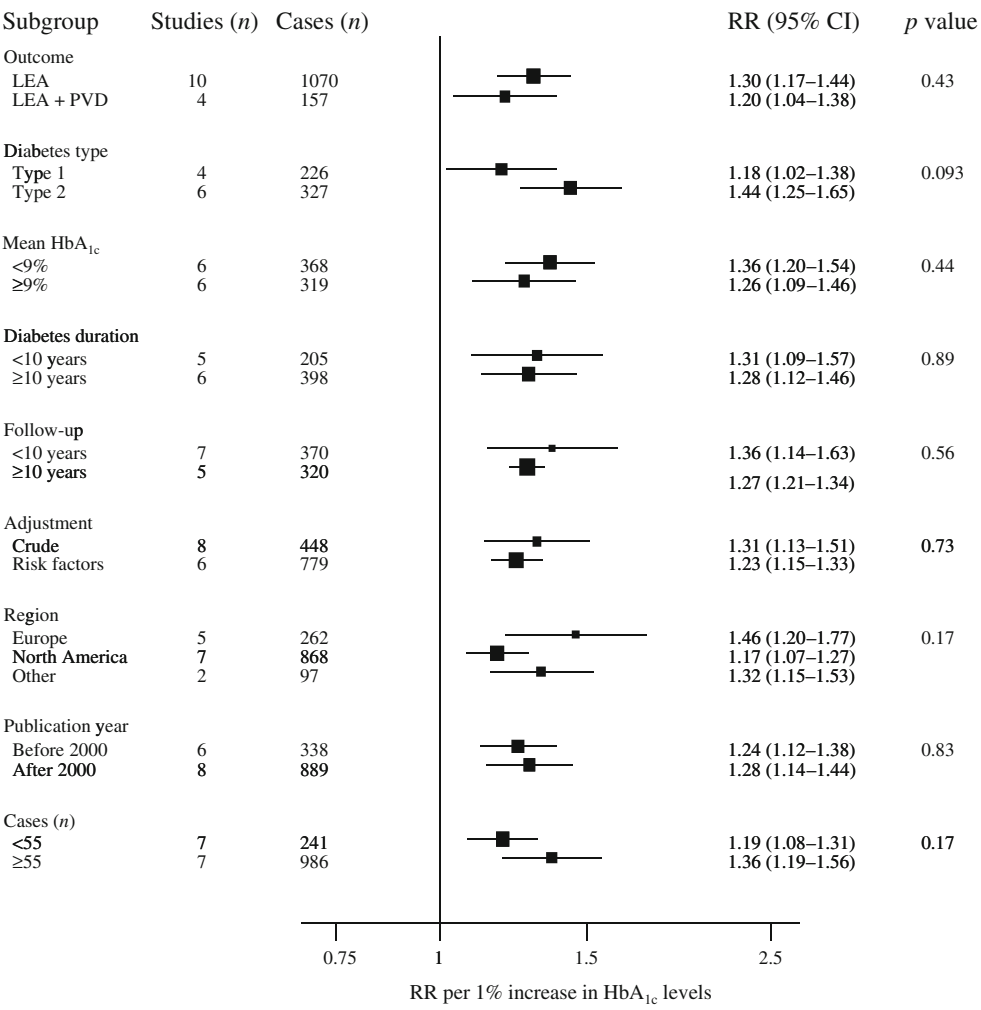

Fig. 5 Association between HbA_{1c} and risk of LEA within subgroups defined by various characteristics. Subgroup risk estimates and heterogeneity *p* values were calculated using random-effects model. The relative risks were not significantly different between studies with higher or lower Newcastle–Ottawa scores (1.21 [95% CI 1.12–1.31] vs 1.37 [95% CI 1.13–1.67], $p=0.43$ for heterogeneity). PVD, peripheral vascular disease

benefit from lowering blood glucose. The results of randomised controlled trials have confirmed this [3, 4] for microvascular complications, while results for macrovascular disease include the possibility of harm [6, 7, 39]. There are limited clinical trial data on the specific effect of glycaemic control on LEA. Trials to date have not been able to demonstrate unequivocally whether improving glycaemic control reduces the risk of LEA. The (DCCT) did not report the effect of glycaemic control on LEA [40] and the UKPDS showed no significant risk reduction associated with randomisation to intensive blood glucose lowering either during the main trial or afterwards [3, 9, 16]. The PROactive trial found no difference in risk of LEA between pioglitazone and placebo groups [17], and in the Kumamoto study no patient in either group had an LEA [5]. Neither the Action to Control Cardiovascular Risk in Diabetes (ACCORD) trial [6] nor the Veteran Affairs Diabetes Trial [41] included LEA in the published primary or secondary outcomes. The Rosiglitazone Evaluated for Cardiac Outcomes and Regulation (RECORD) study included LEA in the primary endpoint, but has not as yet reported how frequently it occurred [18]. The Action in Diabetes and Vascular Disease: Preterax and Diamicron Modified Release Controlled Evaluation (ADVANCE) trial found no difference in the incidence of 'peripheral vascular events' between groups (reduction in relative risk −6 [−9 to 19]) [7]. Although findings from a meta-analysis of clinical trials suggested possible reductions in the risk of peripheral vascular disease with intensified control of blood glucose, these data do not provide conclusive evidence as they were based on a small number of outcomes, of which LEA comprised only a small proportion [39].

Trials to date have either been inadequately powered to find a difference or lowering blood glucose may not in fact lessen the risk of LEA. If patients with higher HbA_{1c} differ fundamentally from those with lower levels in ways that increase their risk of LEA, then lowering blood glucose may not lower the risk of amputation. While use of fenofibrate has recently been shown to lower risk of LEA [42], currently, referral of patients at high risk of amputation to a clinic providing foot care is the most effective preventive measure for major amputation [43].

The 10 year risk of LEA in diabetes varies widely from approximately 1% in Alaskan Natives [44] to 10% in

Barbadians [45] and some UK sites [46]. If lowering blood glucose translates into a lower incidence of LEA, for a population with a 10 year incidence of LEA of 5% [47] and an average HbA_{1c} of 9.5%, an improvement to an average HbA_{1c} of 7.5% would reduce the rate of LEA to roughly 3%, all other things being equal.

Hyperglycaemia may increase the risk of LEA through various mechanisms. It damages tissue via glycation, activates protein kinase C, causes sorbitol to accumulate and increases activity of the hexosamine pathway [48]. This effect manifests as accelerated atherosclerosis and arterial disease, sensory neuropathy, infection and autonomic dysfunction, which deregulates blood flow. Foot deformity, trauma and oedema further contribute to amputation [49]. Improved glycaemic control can potentially modify the risk of sensory neuropathy [5, 50] and possibly the progression of peripheral arterial disease [8]. Other risk factors for LEA include increasing age and duration of diabetes, ethnicity, male sex, renal dysfunction, previous amputation or foot ulceration [34, 51] and, in some studies, smoking [52, 53].

As discussed, the increased risk associated with glycaemia and LEA is likely to be mediated by peripheral vascular disease and peripheral sensory neuropathy. Differences in diagnosis of these conditions may have accounted for some of the heterogeneity we observed. Even if possible, controlling for these factors, which are potentially on the causal pathway to amputation, might lead to statistical over-adjustment with resulting underestimation of the association between glycaemia and LEA [54].

In practical terms, healthcare providers probably already encourage good glycaemic control for patients with diabetes. However, patients may benefit from knowing the magnitude of risk of LEA associated with glycaemia, as LEA is an important complication of diabetes. In the UKPDS, participants rated their decrease in quality of life four times greater for amputation than for blindness in one eye [55].

Regarding potential biases, our inclusion of estimates from studies that adjusted inadequately for confounding (i.e. factors related to both glycaemia and LEA) may have inflated the summary estimate of risk reported by us. However, we found similar overall results for studies reporting crude (or age- and sex-adjusted) or multiply adjusted risk ratios. Misclassification of diabetes type, as in clinical practice, was likely. Yet, misclassification by diabetes type or status, if it occurred equally among those who did and did not have a LEA, is unlikely to have changed our main finding that hyperglycaemia is associated with an increased risk of LEA. However, if patients with (late-onset) type 1 diabetes were more likely to be diagnosed as type 2 diabetes than the reverse, then our study ($p=0.09$) may have missed a real difference in the magnitude of risk associated with glycaemia between type 1 and type 2 diabetes. Another source of misclassification is in the assessment of exposure. Glycated haemoglobin moieties other than HbA_{1c} may have been included in some of the measurements, potentially leading to between-study variations. However, this is also unlikely to have affected the results, as HbA_{1c} is the main component of glycated haemoglobin and most studies stated specifically that they measured HbA_{1c} levels. Due to limited data, we were unable to analyse separately the association between glycaemia and major vs minor amputation. Since LEA is not so much a complication of diabetes as a decision made by a patient advised by surgeons, it is possible that the included studies may not represent usual clinical practice, a possibility reduced by the fact that these studies originated from many areas. The relative risk we report would underestimate the true association between hyperglycaemia and risk of amputation if surgeons were reluctant to operate on chronically ill patients with poor glycaemic control. However, this too is unlikely, since amputation may be a necessary measure to treat an infected/non-healing foot ulcer. In addition, it is difficult to disentangle the contributions of hyperglycaemia and foot ulceration to the risk of amputation, in part because ulceration itself is on the causal pathway to LEA [56]. To diminish the acute effects on HbA_{1c} of immobility and infection, we limited this review to prospective studies of people without acute ulceration or former amputations, but acknowledge that reverse causation may have occurred. Also, we do not know whether our estimates of risk apply to these excluded groups.

In relation to the possible limitations of literature-based meta-analyses, we did not find strong evidence of publication bias, i.e. the increased reporting of smaller studies with positive rather than negative results. Nonetheless, some degree of publication bias may have been present and exaggerated the risk ratios we report. While heterogeneity may limit the generalisability of our findings, it was not accounted for by the clinically relevant characteristics available. Individual-level data are required to assess the shape of the relationship between HbA_{1c} and LEA, or to test differences between other clinical subgroups such as those with or without peripheral arterial disease or sensory neuropathy, or, notably, those with major or minor amputations. There is little reason to believe that glycaemia would increase minor, but not major amputation, or vice versa.

The current review highlights the potential importance of glycaemic control in the prevention of LEA. This study provides an assessment of risk to give to patients. It also provides health economists and planners with estimates to enable them to better model diabetes and its complications. The clinical significance of these observational data is further heightened by the probability that a trial on lowering blood glucose to prevent LEA is unlikely to be done

because: (1) LEA occurs infrequently and would require a very large study; and (2) maintaining differences in glycaemia between groups could be unethical, since lowering blood glucose has already been proven to lower the incidence of other diabetic complications.

In conclusion, the present review shows a strong association between risk of LEA and increased levels of glycaemia in individuals with diabetes. If the association is causal, treatment of glycaemia in patients whose HbA_{1c} remains far above target levels could translate into a large reduction in risk. While amputations occur less frequently than other cardiovascular complications, its consequences may be greater. In the absence of conclusive evidence from clinical trials, and assuming causality, this paper provides further epidemiological support for glucose lowering as a strategy for reducing the risk of LEA; it also provides modellers of diabetes with estimates to more accurately assess the overall burden of hyperglycaemia.

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Duality of interest The authors declare that there is no duality of interest associated with this manuscript.

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