

Dietary Calcium or Vitamin D Intake and Type 2 Diabetes Mellitus Incidence: A Dose-Response Meta-analysis

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Abstract: *Background:* Dietary Calcium and vitamin D intake may have effects on the onset risk of type 2 diabetes mellitus (T2DM), whereas the results of existing studies are inconsistent. We performed a meta-analysis of prospective studies to evaluate the association between dietary calcium or vitamin D intake and the onset risk of T2DM. *Methods:* We searched PubMed, Web of Science, and Embase for relevant studies published through 5 Jan 2021. The relative risks (RRs) with 95% confidence intervals (CIs) of T2DM in relation to dietary calcium or vitamin D intake were pooled with a fix or random-effects model. *Results:* 22 cohort studies were included in this meta-analysis, which comprising 31027 cases and 448578 participants. We found dietary calcium (RR: 0.84; 95% CI: 0.76–0.93) rather than dietary vitamin D (RR: 1.00; 95% CI: 0.92–1.08) reduced the onset risk of T2DM. Subgroup analysis showed the efficacy of dietary calcium intake is only in Asian, women and older population. Dose-response analysis revealed that each 300 mg/day increase in dietary calcium intake reduces the risk of T2DM by 8%. However, the increasing dietary vitamin D intake does not affect the risk of T2DM. *Conclusions:* Dietary calcium intake reduces the onset risk of T2DM but dietary vitamin D has no effect.

Keywords: Calcium, Vitamin D, Type 2 Diabetes Mellitus, Meta-analysis

1. Introduction

Diabetes is one of the most common global chronic diseases with a staggering 463 million cases in 2019, and expected to be 578 million by 2030, 700 million by 2045 [1]. Several large randomized controlled trials (RCTs) demonstrated that dietary interventions could minimize the onset risk of T2DM [2, 3], which could ease the burden of this disease worldwide.

Ca²⁺ is the most abundant ion in the body, which plays both a structural and functional role in bones [4]. Except for its well-known function, it was found that insulin secretion is a calcium-dependent process [5] and also calcium is vital for insulin-mediated intracellular processes in tissues that

responding to insulin, such as muscle and fat [6]. Vitamin D is essential for optimal musculoskeletal health because it promotes calcium absorption, mineralization of osteoid tissue formation in bone, and maintenance of muscle function [7]. Its deficiency has been linked to the onset of T2DM mediated by insulin secretion, insulin sensitivity, and systemic inflammation [8].

Previous reviews have summarized the evidence linking dietary calcium or vitamin D intake with the development of T2DM. However, these reviews were limited by study design, number of participants, and heterogeneity among studies [9, 10]. Here, we conducted a meta-analysis of prospective studies to quantify the effects of dietary vitamin D and

calcium intake on T2DM incidence risk in non-diabetic subjects, as well as to explore the potential of heterogeneity.

2. Materials and Methods

This meta-analysis was conducted according to the Meta-analysis of Observational Studies in Epidemiology (MOOSE) protocol [11]. We registered our study protocol with the International Prospective Register of Systematic Reviews, PROSPERO (registration no: CRD42020182946).

Search strategy. Eligible studies were searched by two investigators (authors L.L. and Q.L.) in PubMed, Web of Science, and Embase from their inception to 5 Jan 2021, using the following search terms: ((((((((((calcium [Title/Abstract]) OR (dairy products [Title/Abstract])) OR (dairy [Title/Abstract])) OR (milk [Title/Abstract])) OR (cheese [Title/Abstract])) OR (yogurt [Title/Abstract])) OR (cream [Title/Abstract])) OR (vitamin D [Title/Abstract])) AND (diabetes [Title/Abstract])) AND (((prospective) OR (follow-up)) OR (cohort)) OR (longitudinal)). In addition, the reference list of the identified articles and relevant reviews and meta-analyses were retrieved manually for additional potential publications.

2.1. Inclusion and Exclusion Criteria

Eligible studies were selected on the basis of following criteria: (1) cohort studies; (2) study subjects were non-diabetic individuals, including those with normal blood glucose and prediabetes at baseline; (3) the exposure was the intake of dietary calcium and/or vitamin D; (4) the outcome was T2DM; (5) the relative risk (RR) or hazard ratio (HR), with the corresponding 95% confidence interval (95% CI) for the risk of T2DM were reported or could be calculated; (6) for the dose-response analysis, the intake of dietary calcium or vitamin D in each category was either reported or could be calculated; (7) the study was published in English. If a study reported several RRs (HRs), the most adjusted ratio for the highest level versus the lowest level was selected to calculate the pooled RR. If the same data were reported in more than one article, we selected the article including the most detailed data. We excluded publications if they were abstracts, meta-analyses, reviews, case-reports, articles without full text, studies involving non-human species, and articles lacking of essential data.

2.2. Data Extraction and Quality Assessment

Data extraction was performed independently by two investigators (authors L.L. and Q.L.): The name of the first author, year of publication, country, geographic location, age, gender, sample size (number of participants and cases), years of follow-up, intake comparison, the corresponding fully adjusted RR (or HR) with 95% CIs and adjusted covariates. The quality of each cohort study was also independently evaluated by investigators mentioned above according to the Newcastle-Ottawa Scale, with a maximum of nine points [12]. Inconsistencies were resolved through group discussions to reach consensus.

2.3. Statistical Analysis

Initially, If $I^2 > 50\%$, the DerSimonian and Laird random-effects model was used; otherwise, the Mantel-Haenszel fixed-effects model was used [13]. The pooled estimates were calculated using the RRs and 95% CIs for all studies. We assumed that HRs reported in the publications were approximately RRs. Any results stratified by gender were treated as independent reports.

Heterogeneity among studies was evaluated by Q test and I^2 statistic. The cut-off points were 25%, 50%, and 75% representing low, moderate, and high degrees of heterogeneity, respectively [14]. To find the probable sources of heterogeneity, meta-regression and subgroup analyses were both performed to evaluate the effect of modifying potential key covariates [15]. Subgroup analyses were performed to assess the potential modifying effects of the following variables on outcomes: geographic location, gender, mean age, follow-up years, baseline characteristics of participants, and adjustment by age, energy, magnesium, calcium, vitamin D or alcohol intake.

A “leave-one-out” sensitivity analysis was used to evaluate whether the overall risk estimate would have been affected by the data of individual study. Publication bias was assessed by the application of Egger’s linear regression test [16] and Begg’s rank correlation test [17].

We used the method developed by Greenland and Longnecker and the available Stata code written by Orsini et al. to estimate dose-response association [18, 19]. We extracted data on intake of dietary calcium or vitamin D in each category (the range or median or mean intake), the number of cases and participants (or person-years) in each category, and the RR (or HR) with 95%CI. The midpoint of the lower and upper categories was regarded as median of the quantile, if the medians or means were not provided. If the highest category with an open upper boundary, we considered the width of that category was the same to the adjacent category. If the lowest category was open-ended, it was assumed to be zero. To evaluate possible non-linear associations between the intake of dietary calcium or vitamin D and the risk of diabetes, restricted cubic splines was used, with 3 knots at fixed percentiles of 10th, 50th, and 90th of the distribution [20]. All analyses were conducted with Stata 14.0 (StatCorp, College Station, TX, USA) and $P < 0.05$ was considered statistically significant.

3. Results

3.1. Search Results and Study Characteristics

As the flow chart showed (Figure 1), a total of 9461 citations (excluding duplicates) from PubMed (n=3539), Web of Science (n=5961), and Embase (n=5817) were initially identified. After reviewing titles and abstracts, we retrieved 130 full-text articles. Ultimately, 22 studies in 15 articles [21-35] were eligible for our inclusion in this meta-analysis. Our meta-analysis included a total of 448578 participants and 31027 cases of T2DM. The details of the 22 studies were

presented in Table 1. The average score of quality assessment was 7.3 (range from 5-9) (Table 3).

Table 1. Characteristics of the 15 articles included in the meta-analysis.

Author, Year	Country	Gender	Age Range (Years)	Participants (Cases)	Follow-up (years)	Exposure	Intake Comparison (Highest vs. Lowest)
Colditz, 1992 (18)	America	F	34-59	84360 (702)	6	Dietary calcium	Q5 vs. Q1 171–740 mg/d
Gagnon, 2011 (19)	Australia	Both	≥25	5200 (199)	5	Dietary calcium	vs. 1060–2317 mg/d
Kim, 2018 (20)	Korea	Both	40-69	8574 (1173)	10	Dietary calcium	Q4 vs. Q1
Kirii, 2009 (21)	Japan	M F	40-69	25877 (634) 33919 (480)	10	Dietary calcium Dietary vitamin D	439 mg/d vs. 348 mg/d 18.0 µg/d vs. 4.7 µg/d
Liu, 2005 (22)	America	F	>45	10066 (805)	8.8	Dietary calcium Dietary vitamin D	1168mg/d vs. 486 mg/d 377 IU/d vs. ≤111 IU/d
Oh, 2017 (23)	Korea	M F	>40	2830 (122) 4669 (168)	6	Dietary calcium	462 mg/d vs. 197 mg/d
Pittas, 2006 (24)	America	F	30-55	83779 (2465) 79232 (4547)	20	Dietary calcium Dietary vitamin D	>1000 mg/d vs. ≤500 mg/d >400 IU/d vs. ≤100 IU/d
Talaei, 2018 (25)	Singapore	Both	45-74	45411 (5207)	11	Dietary calcium	597 mg/d vs. 258 mg/d
van Dam, 2006 (26)	America	F	21-69	41186 (1964)	8	Dietary calcium	661 mg/d vs. 219 mg/d
Villegas, 2009 (27)	China	F	40-70	64191 (2270)	6.9	Dietary calcium	649.6 mg/d vs. 277.5 mg/d
Wu, 2019 (28)	Finland	M F	3-18	556 (31) 578 (19)	31	Dietary calcium	>800 mg/d vs. ≤800 mg/d
Yáñez, 2017 (29)	Sweden, America	F	>18	3414 (169)	12	Dietary calcium	≥1527 mg/d vs. <795mg/d
Abbas, 2014 (30)	Europe	Both	46.5-59.2	27043 (11994)	10.8	Dietary vitamin D	≥6.07µg /d vs. <2.19µg /d
Bao, 2018 (31)	America	F	22-44	8621 (395)	10	Dietary vitamin D	≥400 IU/d vs. <200 IU/d
Eshak, 2018 (32)	Japan	Both	40-79	19168 (494)	5	Dietary vitamin D	13.3 µg /d vs. 3.7 µg/d

Table 1. Continued.

Author, Year	Adjustment for Covariates
Colditz, 1992 (18)	age, BMI, alcohol intake, family history of diabetes, prior weight change, time period, dietary magnesium, dietary potassium
Gagnon, 2011 (19)	N/A
Kim, 2018 (20)	age, sex, residential area, monthly family income, tobacco smoking, alcohol intake, physical activity, and BMI, systolic blood pressure, diastolic blood pressure, and serum creatinine level, serum calcium intake
Kirii, 2009 (21)	N/A
Liu, 2005 (22)	N/A
Oh, 2017 (23)	age, higher education level, regular exercise, FBG level, glycemic load, and dietary magnesium in average total calcium for men; adjusted for age, higher education level, regular exercise, FBG level, and dietary magnesium in average total calcium for women
Pittas, 2006 (24)	age, BMI, hypertension, family history of diabetes, smoking, physical activity, caffeine, alcohol, and state of residence, type of fat, cereal fiber, glycemic load, magnesium, retinol, multivitamin use
Talaei, 2018 (25)	age, sex, dialect, year of interview, and educational level, BMI, physical activity, smoking status, alcohol use, baseline history of self-reported hypertension, and total energy intake, vegetable, fruit, soy-rich pattern and dim sum and meat-rich pattern, coffee, and soda, dietary intake of potassium, magnesium, phosphorus, and vitamin D
van Dam, 2006 (26)	age, total energy intake, BMI, smoking status, strenuous physical activity, alcohol consumption, parental history of diabetes, education level, coffee consumption, sugar-sweetened soft drink consumption, and quintiles of processed meat and other red meat consumption, magnesium intake.
Villegas, 2009 (27)	age, energy intake, BMI, waist-hip ratio, smoking status, alcohol consumption, physical activity, income level, education level, occupation, and hypertension.
Wu, 2019 (28)	age, childhood and adulthood BMI, baseline serum 25OHD levels, parental history of diabetes, fruit and vegetable consumption, physical activity, smoking, and socioeconomic status (parental education years)
Yáñez, 2017 (29)	daily energy intake, maternal age, race/ethnicity, educational attainment, cigarette smoking status, pre-pregnancy BMI, prenatal vitamin use, physical activity, family history of diabetes, alcohol, coffee, sugar-sweetened beverages, red and processed meats, fatty fish, total fiber intake, dietary magnesium and vitamin D intake
Abbas, 2014 (30)	sex, age and study center, total energy, non-fat energy, polyunsaturated fatty acids, monounsaturated fatty acids, saturated fatty acids, physical activity, education, BMI, smoking, alcohol intake
Bao, 2018 (31)	age, parity, race/ethnicity, family history of diabetes, cigarette smoking, alcohol intake, physical activity, estimated ultraviolet B flux, and BMI, total energy intake, intakes of saturated fat, monounsaturated fatty acids, polyunsaturated fatty acids, transunsaturated fatty acids, dietary cholesterol, glycemic load, dietary fiber, magnesium, calcium, vitamin A.
Eshak, 2018 (32)	N/A

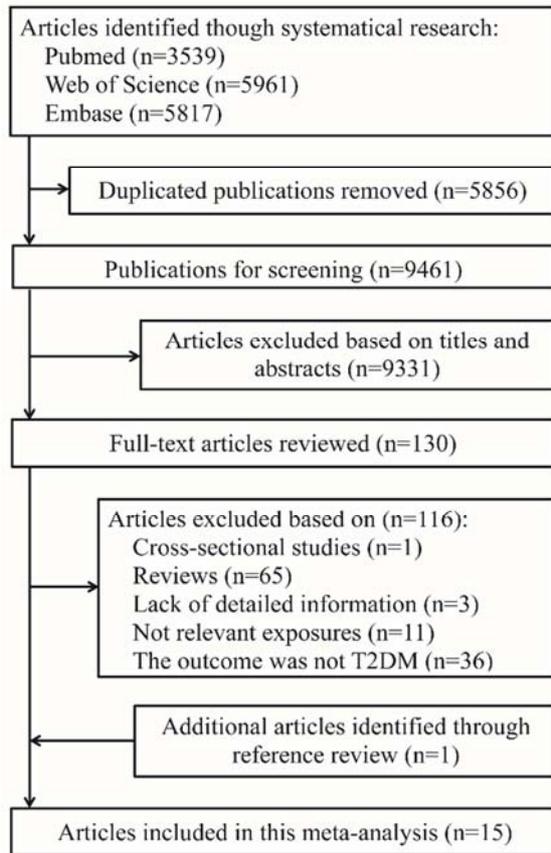
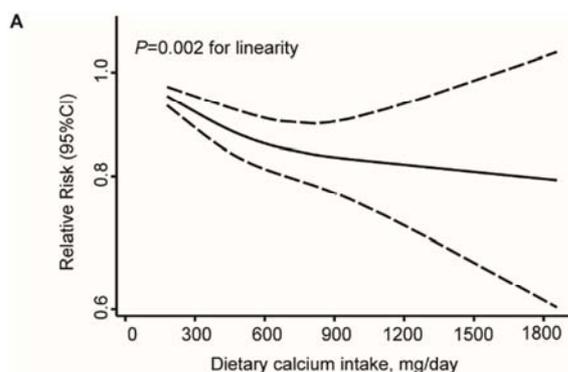


Figure 1. Flow diagram depicting the screening and exclusion of publications included in the meta-analysis.

3.2. Dietary Calcium intake and T2DM

The overall analysis revealed an inverse association between dietary intake of calcium and risk of T2DM based on the random-effects model (RR: 0.84; 95% CI: 0.76–0.93; $I^2=54.7\%$) (Figure 2A). Ten studies provided the required data for dose-response analysis and an inverse linear association between dietary calcium intake and T2DM risk was observed ($P=0.002$). The dose-response analysis also suggested that each 300 mg/d dietary calcium intake increase reduced the risk of T2DM by 8% (RR: 0.92; 95% CI: 0.87–0.97) (Figure 2B). However, no significant non-linear



association was observed by the restricted cubic splines model ($P=0.119$, Figure 3A).

3.3. Dietary Vitamin D intake and T2DM

Consistent with all independent studies included in our analyses, no significant association was found between dietary vitamin D intake and T2DM risk (RR: 1.00; 95% CI: 0.92–1.08; $I^2=0.0\%$) (Figure 4A). The dose-response analysis suggested that no significant association between dietary vitamin D intake and T2DM risk by the per 300 IU/day increase (RR: 1.02, 95% CI, 0.97–1.06; $I^2=19.6\%$) (Figure 4B). No evidence of a non-linear association was found between dietary vitamin D intake and T2DM risk ($P=0.959$ for non-linearity, Figure 3B).

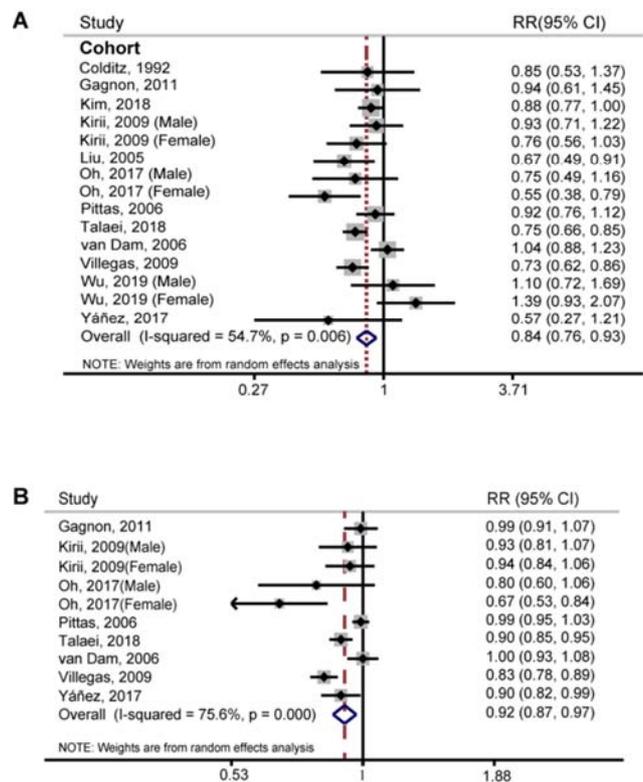


Figure 2. Forest plots summarizing the RR of T2DM for the highest vs. the lowest category of (A) dietary calcium intake and (B) and per 300 mg/day increase in dietary calcium intake. RR: relative risk; CI: confidence interval.

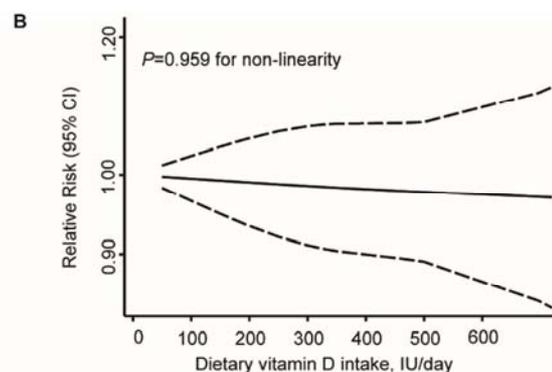
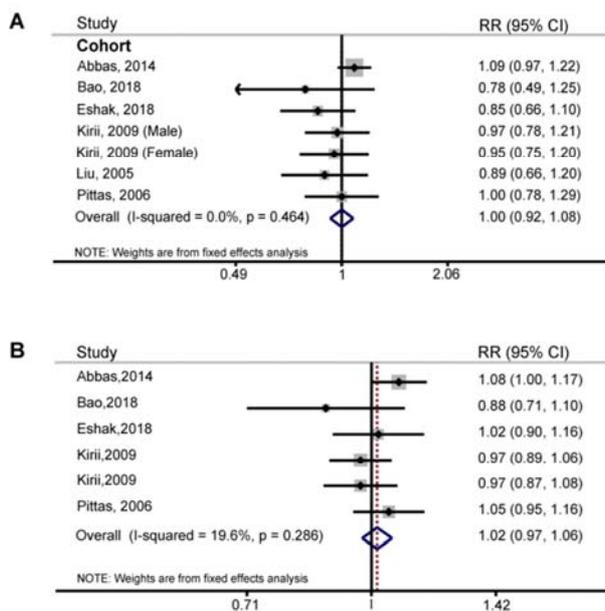


Figure 3. Dose-response analyses of the non-linear association between (A) dietary calcium; (B) dietary vitamin D intake and the risk of T2DM. In each panel, the solid line and dashed lines represent the estimated RR and 95% CI, respectively.

Table 2. Subgroup analyses between dietary calcium intake and the risk of T2DM.

group	N	Cases/controls	RR (95% CI)	I ²	P
Geographic Location					
Asia	7	9862/175835	0.79 (0.73, 0.84)	36.3%	0.152
Europe	3	249/6085	1.14 (0.90, 1.45)	0.0%	0.418
America	5	8033/193682	0.92 (0.82, 1.03)	48.5%	0.101
Gender					
Men	3	787/28476	0.92 (0.75, 1.13)	0.0%	0.466
Women	9	10970/294102	0.82 (0.70, 0.97)	67.6%	0.002
Mean age					
≤50	6	7278/185505	1.00 (0.90, 1.12)	21.3%	0.273
>50	9	10866/190097	0.78 (0.73, 0.84)	27.8%	0.197
Follow-up years					
≥10	8	12364/189970	0.85 (0.79, 0.92)	49.9%	0.051
<10	7	5780/185632	0.78 (0.65, 0.94)	63.5%	0.012
Participants					
Youth	2	50/1084	1.25 (0.93, 1.67)	0.0%	0.432
Pregnancy	1	169/3245	0.57 (0.27, 1.21)	-	-
Healthy adult	12	17925/371273	0.83 (0.78, 0.88)	49.7%	0.025
Adjustment for alcohol					
Yes	7	15686/294365	0.84 (0.75, 0.95)	58.7%	0.024
No	8	2458/81237	0.84 (0.69, 1.02)	57.2%	0.022
Adjustment for magnesium					
Yes	7	12725/231834	0.81 (0.68, 0.96)	63.8%	0.011
No	8	5419/143768	0.86 (0.75, 0.99)	50.9%	0.047
Adjustment for energy intake					
Yes	4	9610/144592	0.81 (0.66, 0.99)	75.8%	0.006
No	11	8534/231010	0.87 (0.80, 0.94)	41.7%	0.071
Adjustment for vitamin D intake					
Yes	2	5376/43449	0.74 (0.66, 0.84)	0.0%	0.479
No	13	12768/332153	0.86 (0.77, 0.96)	53.8%	0.011
Adjustment for age					
Yes	11	16026/302658	0.85 (0.75, 0.96)	63.8%	0.002
No	4	2118/72944	0.81 (0.69, 0.95)	2.2%	0.381

**Figure 4.** Forest plots summarizing the RR of T2DM for the highest vs. the lowest category of (A) dietary vitamin D intake and (B) and per 300 IU/day increase in dietary vitamin D intake. RR: relative risk; CI: confidence interval.

3.4. Heterogeneity and Meta-regression

As for dietary vitamin D intake, there is low study heterogeneity. In contrast, moderate heterogeneity was found in the analysis of dietary calcium intake ($I^2=54.7\%$, $P=0.006$) (Figure 2). Univariate meta-regression analysis on dietary calcium, including covariates of location ($P=0.989$), gender ($P=0.936$), mean age ($P=0.106$), intake of alcohol ($P=0.349$), dietary magnesium ($P=0.149$), vitamin D ($P=0.123$), and energy ($P=0.214$), did not show that any covariation had a significant impact on between-study heterogeneity.

3.5. Subgroup Analyses Stratified by Possible Confounders

Next, we a subgroup analysis was performed on dietary calcium to test the stability of results based on geographic location and gender. The analysis revealed an inverse association in Asia but not in Europe and America, and in women, but not in men. When stratifying studies by mean age in the study, an inverse association was found in the subgroup of ages older than 50 years old, but not in the subgroup of ages younger and equal to 50 (Table 2). In addition, the significant association persisted even after adjusting for major confounding factors, such as intake of dietary

magnesium, vitamin D, total energy, and alcohol were consistent with the overall results of dietary source consumption (Table 2).

As for dietary vitamin D, results of our subgroup analyses

were consistent with the overall results of dietary source consumption (Table 4).

Table 3. Quality assessment of all included studies.

Author, year	Selection			
	Representativeness of cases	Selection of controls	Ascertainment of exposure	No history of outcome
Colditz, 1992 (1)	0	1	1	1
Gagnon, 2011 (2)	0	1	0	1
Kim, 2018 (3)	1	1	1	1
Kirii, 2009 (4)	0	1	1	1
Liu, 2005 (5)	0	1	1	1
Oh, 2017 (6)	0	1	1	1
Pittas, 2006 (7)	0	1	1	1
Talaei, 2018 (8)	0	1	1	1
Van Dam, 2006 (9)	0	1	1	1
Villegas, 2009 (10)	0	1	1	1
Wu, 2019 (11)	0	1	1	1
Yáñez, 2017 (12)	0	1	1	1
Abbas, 2014 (13)	1	1	1	1
Bao, 2018 (14)	0	1	1	1
Eshak, 2018 (15)	1	1	1	1
Average Score: 7.3				

Table 3. Continued.

Author, year	Outcome				Total Score
	Comparability Comparable On confounders	Outcome assessment	Adequate follow-up-time (≥ 5 years)	Follow-up-rate ($> 80\%$)	
Colditz, 1992 (1)	2	1	1	1	8
Gagnon, 2011 (2)	0	1	1	1	5
Kim, 2018 (3)	2	1	1	1	9
Kirii, 2009 (4)	0	0	1	1	5
Liu, 2005 (5)	0	1	1	1	6
Oh, 2017 (6)	1	1	1	1	7
Pittas, 2006 (7)	2	1	1	1	8
Talaei, 2018 (8)	2	1	1	1	8
Van Dam, 2006 (9)	2	1	1	1	8
Villegas, 2009 (10)	1	1	1	1	7
Wu, 2019 (11)	2	1	1	1	8
Yáñez, 2017 (12)	2	1	1	1	8
Abbas, 2014 (13)	1	1	1	1	8
Bao, 2018 (14)	2	1	1	1	8
Eshak, 2018 (15)	0	1	1	1	7
Average Score: 7.3					

Table 4. Subgroup analyses between dietary vitamin D intake and the risk of T2DM.

Subgroup	N	Cases/controls	RR (95%CI)	I ²	P
Geographic Location					
Asia	3	1608/77356	0.93 (0.81, 1.06)	0.0	0.732
Europe	1	11994/15049	1.09 (0.97, 1.22)	-	-
America	3	5747/96719	0.93 (0.78, 1.11)	0.0%	0.626
Gender					
Male	1	634/25243	0.97 (0.78, 1.21)	-	-
Female	4	6227/130158	0.93 (0.81, 1.08)	0.0%	0.810
Adjustment for calcium intake					
yes	1	395/8226	0.78 (0.49, 1.26)		
no	6	18954/180898	1.01 (0.93, 1.09)	0.0%	0.473
Follow-up years					
≥ 10	5	18050/161189	1.03 (0.94, 1.12)	0.0%	0.547
< 10	2	1299/27935	0.87 (0.71, 1.05)	0.0%	0.817
Participants					
Pregnancy	1	395/8226	0.78 (0.49, 1.26)		
Health adult	6	18954/180898	1.01 (0.93, 1.09)	0.0%	0.473
Adjustment for alcohol					
yes	3	16936/102507	1.06 (0.96, 1.17)	2.6%	0.358
no	4	2413/86617	1.06 (0.96, 1.17)	0.0%	0.870
Adjustment for magnesium					

Subgroup	N	Cases/controls	RR (95%CI)	I ²	P
yes	2	4942/87458	0.95 (0.76, 1.18)	0.0%	0.363
no	5	14407/101666	1.01 (0.93, 1.10)	12.0%	0.337
Adjustment for energy intake					
yes	2	12389/23275	1.07 (0.96, 1.20)	45.1%	0.177
no	5	6960/165849	0.94 (0.84, 1.05)	0.0%	0.903
Adjustment for age					
yes	3	16936/102507	1.06 (0.96, 1.17)	2.6%	0.358
no	4	2413/86617	0.92 (0.81, 1.04)	0.0%	0.870

RR (95%CI): risk estimate and 95% confidence interval; P: value for heterogeneity within each subgroup

3.6. Publication Bias and Sensitivity Analysis

Our sensitivity analysis indicated that there was no significant change when any one study was omitted at a time for dietary calcium and vitamin D (Figure 5). We then looked for publication bias using Begg's funnel plots and Egger's test. As for dietary calcium, result revealed no significant

publication bias based on Egger's test ($P=0.966$) (Figure 6A). With respect to dietary vitamin D, publication bias was statistically significant ($P=0.003$) (Figure 6B). We found that result displayed no significant publication bias ($P=0.113$) after excluding the study performed by Abbas *et al.* (30), which used 24-h diet recall data, and most probably, such an exposure assessment might not be accepted for publication today.

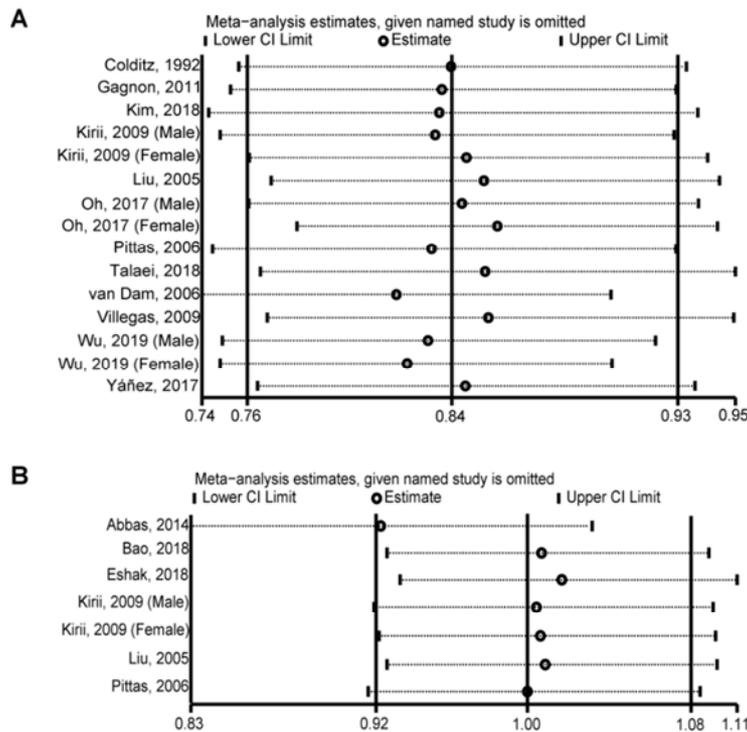


Figure 5. Analysis of influence of individual study on the association between (A) dietary calcium; (B) dietary vitamin D and the risk of T2DM. Open circle indicates the pooled RR, given named study is omitted. Horizontal lines represent the 95% CI.

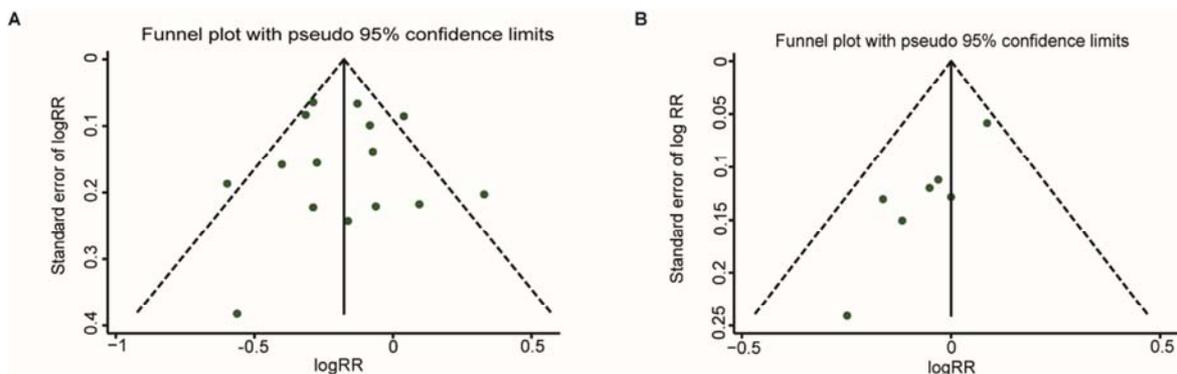


Figure 6. Funnel plot of (A) dietary calcium; (B) dietary vitamin D and the risk of T2DM.

4. Discussion

The meta-analysis presented in this paper was based on 22 published studies including 448578 participants and 31027 cases of T2DM. We found an inverse association between T2DM risk and dietary calcium intake. Moreover, our dose-response analysis revealed that each 300 mg/day increase in dietary calcium intake reduces T2DM risk by 8%. On the other hand, we found no significant association between dietary vitamin D intake and T2DM risk.

As to the correlation between dietary calcium intake and T2DM risk, a previous meta-analysis concluded that dietary calcium intake was not independently associated with T2DM risk and that the inverse association between dietary calcium intake and T2DM risk in prior observational studies may be influenced by magnesium intake [9]. However, in our analysis, after adjusted for dietary magnesium, an inverse correlation between dietary calcium intake and T2DM risk still exists. The reason for the different results may be that our meta-analysis included more studies (15 vs. 7) and incident cases (18144 vs. 11195) than the previous meta-analysis. Nevertheless, we found that inverse relationship was only observed in Asia, but not in America and Europe. Racial differences in calcium metabolism may explain the result, for example, dietary calcium is likely utilized more efficiently in Chinese than Westerners as reported [36]. Moreover, a subgroup analysis revealed that this inverse correlation was present in women but not in men. The gender disparity could be ascribed to sex hormones. However, most women in our study were post-menopausal, and the inverse association also exists in subgroup of age above 50, possibly there were other explanations related to lifestyle. Women were inclined to consumed more calcium and dairy products than men [24]. On the other hand, smoking and alcohol consumption, which were common among males, may influence calcium metabolism *via* decline of the vitamin D parathyroid hormone system among smokers [37] or by reduction of bone mineral density in alcohol drinkers [38]. Additionally, post-menopausal women were calcium deficiency [39]. In our subgroup analyses, alcohol was identified as a confounder. As only two studies were adjusted for vitamin D intake, it was unclear whether the protective effect of dietary calcium was independent or depending on vitamin D.

Different from dietary calcium, our analysis suggests that there is no association between dietary vitamin D intake and T2DM risk. It is reported that vitamin D can affect glucose homeostasis such as insulin secretion, insulin sensitivity and systemic inflammation [8]. Animal studies and human observational studies linked vitamin D deficiency to numerous chronic diseases including hypertension, and diabetes [7, 40]. However, multiple RCTs have failed to demonstrate significant benefits of vitamin D supplementation for diabetes. It is possible that many prior epidemiological associations between vitamin D deficiency and chronic diseases were caused by unmeasured residual confounding. For many of the chronic diseases previously relation to vitamin D deficiency, their preclinical stages likely lead to the

vitamin D deficiency that subsequently cause false associations in some observational studies [41], so our results demonstrate that dietary vitamin D may have no protective effect on T2DM in people with normal vitamin D levels at baseline.

Our meta-analysis has several strengths. First, this is the first systemic dose-response meta-analysis that quantifies the association between the intake of dietary calcium or vitamin D and T2DM risk. Second, our meta-analysis included prospective studies from large populations and long-term follow-up studies, and thus increases the statistical power by detecting modest associations. Third, we performed several subgroup analyses to identify the potential sources of heterogeneity and to evaluate the robustness of our results. We found difference between subgroups of mean ages, geographical areas and gender-specific difference in the effects of dietary calcium intake on the risk of T2DM. Lastly, we included the studies which used estimates from the fully adjusted models to minimize the effects of potential confounding factors.

Our analysis has limitations. First, the possibility of residual confounding still exists, given the observational nature of the studies. Second, inaccurate measurement seemed inevitable and might weaken true associations, because the consumption of various dietary factors was assessed by food frequency questionnaires in almost all of cohort studies. Third, a number of included studies did not adjust the vitamin D information when assessing the relationship between dietary calcium intake and the outcome. It is also not clear whether the protective effect of dietary calcium on T2DM is independent or not. Finally, there was some evidence of publication bias as for dietary vitamin D. We cannot completely rule out the possibility that it affected the significance of our results.

Large-scale epidemiological studies, in particular prospective cohort studies that adjust for vitamin D, calcium intake or other possible covariates separately should be conducted in order to further test the association between dietary calcium or vitamin D intake and the risk of T2DM. Additionally, researches are needed to examine the effects of dietary calcium and/or vitamin D intake on indexes such as glucose tolerance, insulin secretion, etc. to improve homeostasis rates of glucose and insulin among non-diabetic subjects.

5. Conclusions

Our meta-analysis indicates that dietary calcium intake is inversely correlated with T2DM risk and dose-response analysis suggests that for each 300 mg/d increase in dietary calcium intake reduces the risk of T2DM by 8%. The inverse association is only found in Asia but not Europe and America, and in women, but not in men. In contrast, we found no correlation between dietary vitamin D intake and the risk of T2DM. Our results strongly support the general notion that sufficient calcium are protective factors for T2DM incidence.

The dose-response analyses also consist with these findings. These results may have important public health implications in the prevention of T2DM incidence.

Availability of Data and Materials

All data generated or analyzed during the study are included in this published article.

Authors' Contributions

L.L. and Q.L. designed the study; L.L. and Q.L. assessed the studies for inclusion, extracted the data, and assessed the quality of the included studies; L.L. and Q.L. conducted the meta-analysis; other authors tabulated the data and checked the details of all the files; L.L. wrote the first draft of the manuscript; and J.W. and H.W. provided critical input for the manuscript. Y.X., X.F., S.Z. and M.Z. participated in the helpful discussion and manuscript revision. Y.C. participated in the English grammar revision of the manuscript. W.Y. did critical revision of the manuscript for important intellectual content and study supervision. All authors have contributed significantly, and all authors are in agreement with respect to the content of the manuscript.

Conflict of Interest

The authors declare that they have no competing interests.

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