Antioxidant Micronutrients and Cardiovascular Risk in Patients with Diabetes: A Systematic Review

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Abstract

Background: **Inverse associations between micronutrient intake and cardiovascular outcomes have been previously shown, but did not focus on diabetic patients.**

Objective: **To systematically review the role of micronutrients in the development/presence of cardiovascular outcomes in patients with diabetes.**

Methods: **We searched Medline, Embase, and Scopus (January/1949–March/2012) for observational studies that evaluated micronutrients and cardiovascular outcomes in patients with diabetes, and then selected and extracted the data (two independent reviewers).**

Results: **From the 15 658 studies identified, five were included, comprising three case-control and two cohorts, with a follow-up of 7–15 years. A meta-analysis was not performed due to the different antioxidant micronutrients (types and measurement methods) and outcomes evaluated. The micronutrients assessed were vitamin C intake in diet and/or supplementation, chromium and selenium in toenail samples, and α-tocopherol and zinc in serum levels. Intake of > 300 mg of vitamin C through supplementation was associated with increased risk of cardiovascular disease, coronary artery disease (CAD), and stroke (RR 1.69–2.37). High levels of α-tocopherol in serum were associated with 30% lower CAD risk in another study (HR 0.71; 95%CI 0.53–0.94). Among minerals (zinc, selenium, and chromium), an inverse association between zinc and CAD was observed; levels lower than 14.1 μmol/L were associated with an increased risk for CAD (RR 1.70; 95%CI 1.21–2.38).**

Conclusion: **The information available on this issue is scarce. Further prospective studies are needed to elucidate the role of these nutrients in the cardiovascular risk of patients with diabetes. (Arq Bras Cardiol. 2013; [online].ahead print, PP.0-0)**

Keywords: **Micronutrients; Antioxidants; Risk Factors; Cardiovascular Diseases; Diabetes Mellitus.**

Introduction

Biological research supports a key role for oxidative stress in atherogenesis; free radical-mediated damage induces oxidative changes in low-density cholesterol particles that initiate and promote atherosclerotic changes. This process could be reversed or prevented by the use of antioxidants[1]. Observational studies have shown inverse associations between antioxidant intake and cardiovascular events[2], but randomized clinical trials have not shown any benefit of antioxidants in cardiovascular events[3]. However, these studies did not focus on patients with diabetes mellitus (DM), a population with high cardiovascular disease risk.

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Coronary and cerebrovascular diseases, which are caused primarily by atherosclerosis, are the major cause of morbidity and mortality in patients with DM[4], and occur more frequently and more severely in these patients[5]. Hyperglycemia in DM is characterized by a high oxidative stress state[6], which is closely related to the genesis of chronic complications of diabetes, including cardiovascular diseases[7]. Particular attention has been given to the applicability of antioxidant therapy (endogenous enzymes and dietary substances) in the prevention and management of diabetic complications, especially atherosclerotic cardiovascular disease[8].

Lifestyle changes and a healthy diet are included in the prevention and/or treatment of atherosclerosis in patients with or without DM. High intake of fruits, vegetables, whole grains, and oil seeds and low intake of sodium are usually recommended[9]. Epidemiological studies have shown that certain foods with antioxidant properties are associated with a reduction in inflammatory markers and low-density cholesterol oxidation[8], and consequently, improved endothelial function[10].

The aim of this study was to systematically review the role of vitamins (vitamins A, C, and E) and minerals (zinc, selenium, chromium, manganese, and copper) with antioxidants properties in the presence or development of clinical cardiovascular outcomes in patients with DM.

Methods

Literature search

The search was performed to select observational studies that evaluated the role of antioxidant micronutrient intake (vitamins and minerals) in the presence or development of cardiovascular events in patients with DM. The databases used in the search were *Medline* from *Pubmed, Embase,* and *Scopus* for the period from January 1949 to March 2012. The search strategy included terms referring to antioxidant micronutrients: *"micronutrients," "antioxidant micronutrient," "trace elements," "biometals," "antioxidants," "vitamins," "antioxidant vitamins," "vitamin C," 'ascorbic acid," 'vitamin E," "tocopherols," "alfa-tocopherol," "β-carotene," "vitamin A," "pro-vitamin A," "minerals," "antioxidant minerals," "diet," "diet therapy," "zinc," "copper," "manganese," "chromium," "selenium"*, to patients (type 1 or type 2 DM): *"Diabetes Mellitus, Type 1," "Diabetes Mellitus, Insulin-Dependent," "Diabetes Mellitus, Juvenile-Onset," "Diabetes Mellitus, Sudden-Onset," "Diabetes Mellitus, Type I," "IDDM," "Diabetes Mellitus, Brittle," "Diabetes Mellitus, Ketosis-Prone," Autoimmune Diabetes, "Diabetes Mellitus, Type 2," "Diabetes Mellitus, Ketosis-Resistant," "Diabetes Mellitus, Non-Insulin-Dependent," "Diabetes Mellitus, Slow-Onset," Stable Diabetes Mellitus, "Diabetes Mellitus, Type II," NIDDM, "Diabetes Mellitus, Adult-Onset," "Diabetes Mellitus, Noninsulin Dependent," "Maturity-Onset Diabetes Mellitus,"* and type of study (observational), using a previously validated list of terms available at: http://www.sign.ac.uk/methodology/filters.html#obs.

The search strategy described above was used to identify studies on *Pubmed*. Similar terms were searched for in other databases. There was no restriction of the language used in the publications. The article references included in this review were consulted to identify other potentially eligible studies.

Inclusion and exclusion criteria

We included observational studies (case-control studies and cohorts irrespective of their prospective or retrospective nature) that evaluated the role of antioxidant micronutrient intake (from diet and/or supplements) in the presence or development of major cardiovascular events such as myocardial infarction or revascularization, stroke, sudden death, and death from cardiovascular causes in patients with type 1 or type 2 DM.

In selecting the studies, the antioxidant micronutrients looked at were vitamin A (beta-carotene), vitamin C (ascorbic acid), vitamin E (tocopherol), zinc, selenium, chromium, manganese, and copper. The outcomes considered were major cardiovascular events (cardiovascular death, stroke, myocardial infarction, and myocardial revascularization) and their individual components (fatal and non-fatal myocardial infarction, fatal and non-fatal stroke, sudden death or myocardial revascularization).

Study selection and data extraction

Two reviewers (R.A.S. and F.M.S.) independently reviewed the titles and abstracts of each article identified in the literature search. In this first stage all articles that clearly did not meet the inclusion criteria were rejected. The selected articles were analyzed by reading the full text, and the eligible articles were then identified. Disagreements between reviewers at this stage of article analysis were resolved by discussion. The concordance, estimated by Kappa coefficient, was good (Kappa = 0.79).

Data extraction from each study included in this review was conducted independently by two reviewers (R.A.S. and F.M.S.) using a standardized instrument. The data extracted were publication identification, study design, sample size, follow-up duration (in cohort studies), and participants' general characteristics (type of DM, age, gender, body mass index, diabetes treatment, hypertension, and smoking). The data on diet characteristics and micronutrient antioxidants evaluated (quantity, measurement unit, and assessment method) were also extracted. The data extracted concerning cardiovascular outcomes were event type, case numbers, and the estimated risk as presented in the manuscript [relative risk (RR), odds ratio (OR), or hazard ratio (HR)]. We extracted the risk estimate data that considered the largest number of covariates in the analyses.

Quality assessment of studies

The methodological quality of each study included in this review was assessed independently by two reviewers (R.A.S. and F.M.S.) from a questionnaire developed by the authors. The questionnaire was based on four instruments for quality assessment of observational studies designed by the Scottish Intercollegiate Guidelines Network and Critical Appraisal Skills Programme, as proposed in the Cochrane Handbook[11]. The questionnaire included issues related to the study aim (clarity and specificity), the inclusion and exclusion criteria used to select the participants, sample size of groups, number of patients lost from each group, assessment form of exposure status to the factor studied, and outcomes (if standardized assessment was made by blinded investigators as to the participant exposure status).

Results

Literature search

From the 15 658 articles identified, 2865 were excluded because they were duplicated among the databases searched. After analysis of titles and abstracts, 12 766 articles were excluded because they did not meet the inclusion criteria and 27 articles were selected for reading the full text. After evaluating the full texts, 22 articles were excluded because of the following criteria: five studies were not observational,

one study involved patients with pre-diabetes and without diabetes, three studies did not assess antioxidant micronutrient effect, seven studies did not evaluate cardiovascular outcomes, and six studies included DM as a covariate in estimating cardiovascular risk and sub-analyses did not include only patients with DM. New articles were not identified from the reference lists of studies consulted. Therefore, five studies were included in this review[12-16]. The study selection flow diagram is shown in Figure 1.

General characteristics of the studies

The main characteristics of the five studies included are described in Table 1. Three of them presented a case-control design[12,14,15] and two were cohort studies[13,16] with a follow-up ranging from 7[16] to 15 years[13]. One study was conducted in patients with type 1 DM[15], one study included patients with type 2 DM[16], and in one manuscript the authors reported that the majority of the participants had type 2 DM[13]. The other two studies did not specify the type of diabetes[12,14]. Sample size ranged from 121[15] to 1923 participants[13]. The age of the patients ranged from 34 to 75 years. Two studies included both men and women[15,16], two studies were performed only in men[12,14], and one study was conducted only in women[13]. Two studies described the treatment of DM; in one of them most of the participants were using oral antidiabetic agents[16]; whereas in the other study, approximately 70% of patients were using insulin and/or oral antidiabetic agents[13]. Only two studies reported the number of hypertensive participants, current smokers, and the waist-to-hip ratio values[13,15].

Different antioxidant micronutrients were evaluated in the studies and different methods were used to measure them. Vitamin C provided in dietary intake and/or supplementation was assessed by food frequency questionnaire[13], chromium and selenium were quantified in samples[12,14], and α-tocopherol and zinc were measured in serum[15,16]. The usual diet composition was not described in any study, only a partial dietary description of saturated fatty acids, vitamin E, and beta-carotene was reported in one study[13].

Cardiovascular outcomes were differently evaluated among the studies: two studies evaluated the presence of cardiovascular disease[12,14], two reported the presence of coronary artery disease[15,16] and one other study reported mortality by cardiovascular disease, coronary heart disease, and stroke[13]. Because of these differences we could not perform a meta-analysis of the data extracted. Therefore, the main results of each study included in this review are shown in Table 2 and discussed.

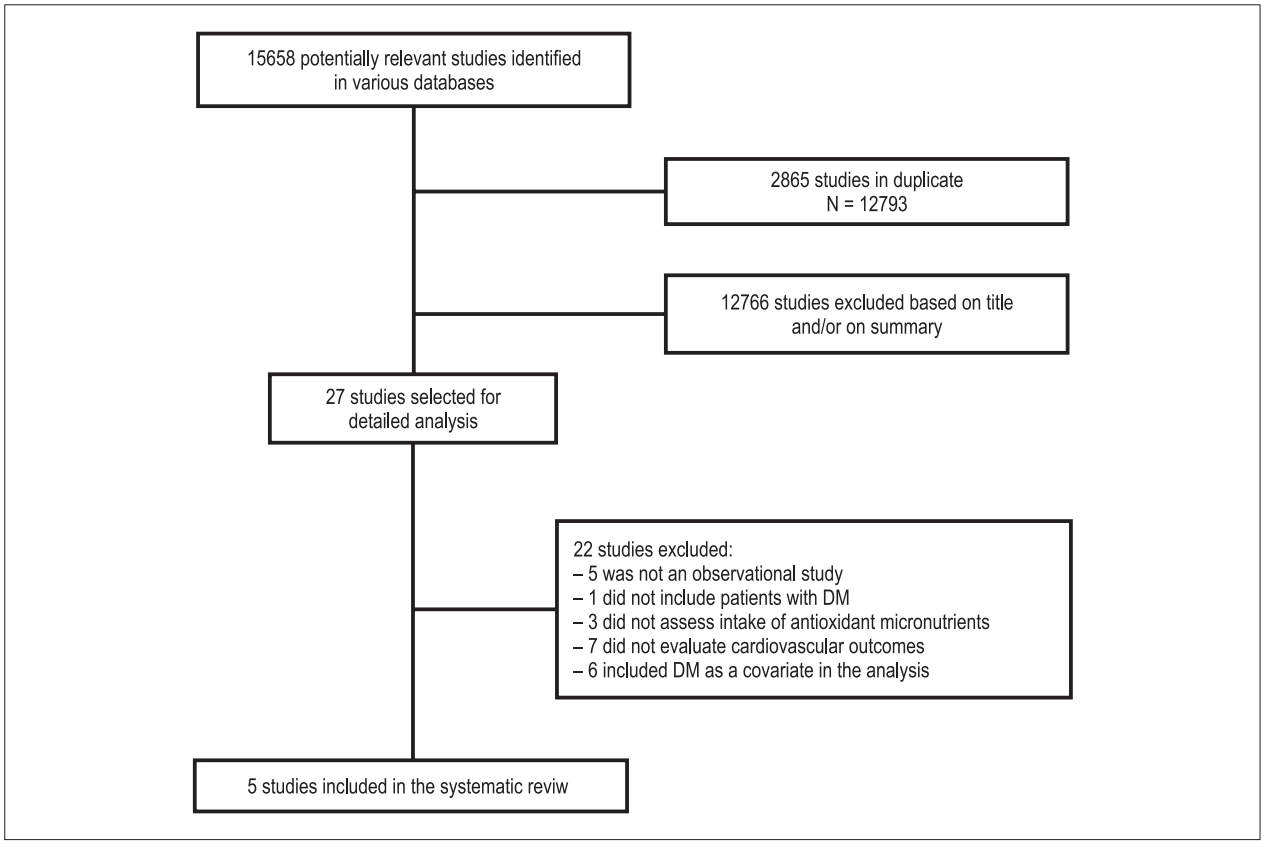

Figure 1 - *The study selection flow diagram.*

Table 1 - Main features of studies

Author, year	Design (follow-up)	n	DM	DM duration	Age (years)	Gender	BMI (kg/m²)	Micronutrient
Rajpathak et al[12] (2004)	case-control	886	not reported	not reported	40–75	100% men	not reported	chromium
Lee et al[13] (2004)	cohort (15 years)	1923	not reported	10.3 years	62.2	100% women	30.1	vitamin C
Rajpathak et al[14] (2005)	case-control	886	not reported	not reported	40–75	100% men	not reported	selenium
Costacou et al[15] (2006)	case-control	121	type 1	26.7 years	34.6	47.9% women	24.2	α-tocopherol
Soinio et al[16] (2007)	cohort (7 years)	1059	type 2	not reported	45–64	45.1% women	27.9	zinc

DM: Diabetes Mellitus; BMI: body mass index.

Table 2 - Main results of the studies included in the review

Author (year)	Micronutrient (measure unit)		Statistical analysis criteria	Outcome (number of cases /total number)	RR/OR/HR (CI 95%)	Variables considered for adjustment in multivariate analysis
Rajpathak et al. (2004)[12]	toenail chrome (µg/g)		upper quartile (>2.08) vs. other quartiles	CVD (198/886)	OR = 0.68 (0.42–1.10)	Age, BMI, alcohol, smoking, family history of AMI, physical activity, hypercholesterolemia, hypertension, dietary fats, fiber, glycemic load, folate and selenium levels, and mercury in toenail.
Lee et al. (2004)[13]	vitamin C (mg/day)	diet and supplementation	upper quintile (>667) vs. other quintiles	CVD (281/1923)	RR = 1.84 (1.12–3.01)	Age, energy, WHR, BMI, physical activity, smoking, alcoholism, education, marital status, HRT, treatment and duration of DM, dietary fats, vitamin E, β-carotene and folate.
				CAD (175/1923)	RR = 1.91 (1.05–3.48)	
				Stroke (57/1923)	RR = 2.57 (0.86–7.66)	
		only diet	upper quintile (>251) vs. other quintiles	CVD (281/1923)	RR = 1.11 (0.66–1.87)	Age, energy, WHR, BMI, physical activity, smoking, alcoholism, education, marital status, HRT, treatment and DM duration, dietary fats, vitamin E, β-carotene, folate, and vitamin C supplements.
				CAD (175/1923)	RR = 1.08 (0.57–2.06)	
				Stroke (57/1923)	RR = 1.89 (0.60–6.03)	
		only supplementation	upper quartile (>300) vs. other quartiles	CVD (281/1923)	RR = 1.69 (1.09–2.44)	Age, energy, WHR, BMI, physical activity, smoking, alcoholism, education, marital status, HRT treatment and DM duration, dietary fats, vitamin E, β-carotene, folate, and vitamin C.
				CAD (175/1923)	RR = 2.07 (1.27–3.38)	
				Stroke (57/1923)	RR = 2.37 (1.01–5.57)	
Rajpathak et al. (2005)[14]	toenail selenium (µg/g)		upper quartile (>1.20) vs. other quartiles	CVD (198/886)	OR = 1.47 (0.92–2.35)	Age, BMI, alcohol, smoking, family history of MI, physical activity, hypercholesterolemia, hypertension, dietary fats, fiber, glycemic load, folate and chromium, and mercury levels in toenail.
Costacou et al. (2006)[15]	serum α-tocopherol (µg/ml)		high levels (>10.45) vs. low levels	CAD (54/121)	HR = 0.71 (0.53–0.94)	Adjustment model is not specified.
Soinio et al. (2007)[16]	serum zinc (µmol/L)		lower quartile (<14.1) vs. other quartiles	Fatal CAD (156/1059)	RR = 1.70 (1.21–2.38)	Age, sex, DM duration, total cholesterol, HDL-c, triglycerides, HbA1c, GFR, hypertension, smoking, BMI, residence place, and DM treatment.
				Fatal CAD or non-fatal AMI (254/1059)	RR = 1.37 (1.03–1.82)	

CAD: coronary artery disease; CVD: cardiovascular disease; DM: Diabetes Mellitus; AMI: acute myocardial infarction; OR: odds ratio; HR: hazard ratio; RR: relative risk; CI: confidence interval; WHR: waist-to-hip ratio; BMI: body mass index; HRT: hormone replacement therapy; HDL-c: HDL-cholesterol; HbA1c: glycated hemoglobin; GFR: glomerular filtration rate.

Main findings of the studies

Antioxidant vitamins and cardiovascular outcomes

The role of antioxidant vitamins in cardiovascular disease development was evaluated in two studies[13,15].

The relationship between vitamin C intake (assessed by a food frequency questionnaire validated in a subsample of the study population) and cardiovascular outcomes in postmenopausal women with DM was evaluated in a prospective cohort study followed for 15 years[13]. Cardiovascular outcomes (cardiovascular disease mortality, coronary heart disease and stroke) were defined based on the International Classification of Diseases: the codes potentially related to the diagnoses of interest were selected according to the description in the records of local deaths (Iowa, USA). Vitamin C intake of more than 667 mg/day (diet and/or by supplementation) approximately doubled the risk of mortality from cardiovascular disease and coronary artery disease in patients with diabetes. When dietary and supplemental vitamin C were analyzed separately, only supplemental vitamin C showed a positive association with mortality endpoints: the use of at least 300 mg/day of vitamin C supplements was associated with higher risk of cardiovascular disease mortality (RR 1.69; 95%CI 1.09–2.44), coronary artery disease (RR 2.07; 95%CI 1.27–3.38) and stroke (RR 2.3; 95%CI 1.01–5.57) than using smaller quantities of supplementation.

The effect of α-tocopherol, γ-tocopherol, and retinol on the incidence of coronary artery disease in patients with type 1 DM was evaluated in a study of 54 cases and 67 controls derived from a cohort study conducted in Pittsburgh, USA[15]. Cases were defined by the participants who first developed coronary artery disease, as determined by one of the following criteria: physician-diagnosed angina, myocardial infarction confirmed by Q-waves on electrocardiogram, hospital records (Minnesota code 1.1 or 1.2), angiographic stenosis ≥50%, coronary artery bypass surgery, angioplasty, or ischemic electrocardiographic changes during the follow-up period. Serum levels of α-tocopherol ≥10.45 μg/ml were inversely associated with coronary artery disease (HR 0.71; 95%CI 0.53-0.94). However, when multivitamin supplement users were compared to nonusers, the protective effect of this micronutrient was observed only among supplement users (HR 0.22; 95%CI 0.10–0.49). It is noteworthy that the authors did not specifically report the type of supplement used by the study participants.

Antioxidant minerals and cardiovascular risk

The possible association between zinc, chromium and selenium and presence or development of cardiovascular events in patients with DM were evaluated by three studies[12,14,16].

A cohort study with 7 years of follow-up investigated serum zinc levels as a predictor of coronary artery disease in 1050 patients with type 2 DM from Finland[16]. The outcomes evaluated were mortality from coronary artery disease based on medical records and death certificates, and myocardial infarction incidence according to the World Health Organization criteria (chest pain, enzyme changes and electrocardiogram). Patients with ≤ 14.1 μmol/L of serum zinc at baseline were at higher risk of death from coronary artery disease (RR 1.7; 95%CI 1.21–2.38) and fatal and non-fatal myocardial infarction (RR 1.37; 95%CI 1.03–1.82) than patients with serum levels ≥14.1 μmol/L.

In a case-control study (derived from the *Health Professionals Follow-up Study*), toenail levels of chromium[12] or selenium[14] were determined in 198 male patients with DM and prior cardiovascular disease, as well as 688 male patients with DM and without cardiovascular disease. Cardiovascular disease was considered present when subjects presented fatal or non-fatal myocardial infarction as defined by the World Health Organization criteria, coronary artery bypass grafting, angioplasty or stroke. In a multivariate analysis adjusted for the other potential confounding factors, there was no association between chromium[12] or selenium[14] levels and cardiovascular outcomes.

Quality evaluation

Quality evaluation is shown in Table 3. None of the studies included meets all the criteria previously established to evaluate methodological quality. However, all five studies were for the purpose of answering a clear and focused question and four of them assessed the exposure status and outcomes in a standardized and valid method. The information regarding the outcomes was collected from population-based registries in the two cohort studies selected[13,16]. In the three case-control studies[12,14,15], outcomes were measured in a valid and standardized way. Potential confounding factors were considered in analysis of data from four studies[12-14,16]. Only one study showed no clear results, because the authors did not describe which covariates were used for multivariate regression adjustment[15]. Among the three case-control studies, none of them described whether the follow-up losses were similar between the groups[12,14,15]. In the two cohort studies[13,16], follow up duration was considered appropriate and the selection of participants was controlled for potential confounding factors.

Discussion

The purpose of this systematic review was to evaluate the role of antioxidant micronutrients in the presence or development of cardiovascular events in patients with DM. However, the high clinical heterogeneity among the studies obtained hindered the performance of a meta-analysis. Moreover, information on this issue is scarce and of low quality. Vitamin C, vitamin E (α-tocopherol), zinc, selenium, and chromium were micronutrients with antioxidant properties evaluated by the case-control and cohort studies included in this review. The outcomes analyzed were myocardial infarction, stroke, myocardial revascularization, sudden death, and death from cardiovascular causes.

The use of more than 300 mg/day of vitamin C by supplementation was associated with increased cardiovascular risk[13]. Interestingly, this is not what is

Table 3 - Methodological quality of studies included in this review

	Cohort studies		Case-control studies		
	Lee et al. (2004)[13]	Soinio et al. (2007)[16]	Rajpathak et al. (2004)[12]	Rajpathak et al. (2005)[14]	Costacou et al. (2006)[15]
Items related to all observational studies					
Issue clear, focused, and appropriate	Yes	Yes	Yes	Yes	Yes
Exposure status assessed by valid and standardized way	Yes	Yes	Yes	Yes	No
Outcomes assessed by valid and standardized way	Yes	Yes	Yes	Yes	Yes
Outcomes evaluated by investigators blinded to the exposure	Not described	Not described	Yes	Yes	Not described
Potential confounding factors considered in the analysis of data	Yes	Yes	Yes	Yes	Not described
Results clearly presented and discussed	Yes	Yes	Yes	Yes	No
Items related to cohort studies					
Sufficient follow-up duration	Yes	Yes	Not applicable	Not applicable	Not applicable
Selection of participants controlled for potential confounders	Yes	Yes	Not applicable	Not applicable	Not applicable
Items related to case-control studies					
Sample size similar between cases and controls	Not applicable	Not applicable	No	No	Yes
Data collected similarly for cases and controls	Not applicable	Not applicable	Yes	Yes	Yes
Exclusion criteria applied similarly for cases and controls	Not applicable	Not applicable	Not described	Not described	Not described
Clearly defined cases	Not applicable	Not applicable	Yes	Yes	Yes
Controls clearly defined	Not applicable	Not applicable	Yes	Yes	Yes
Follow-up losses similar between cases and controls	Not applicable	Not applicable	Not described	Not described	Not described

reported for subjects without diabetes[2,17]. A systematic review of 15 cohort studies with 374,488 subjects without DM showed an inverse association between higher intake of vitamin C (diet and supplement) and risk of coronary artery disease (RR 0.84; 95%CI 0.73–0.95)[2], but the results were not confirmed with the use of supplemental vitamin C only in the same study[2]. In clinical trials with long follow-up periods analyzed in other reviews, vitamin C supplement use had no significant effect on the risk of myocardial infarction and stroke in subjects without diabetes[17]. The inconsistency of these findings may be partially explained by the presence of diabetes and the recommended daily intake of the vitamin. Vitamin C can act as a pro-oxidant interacting with free iron[18] and among patients with DM an iron metabolism disorder seems to occur, with an increase in free iron stores[19]. Alternatively, vitamin C could have promoted protein glycation[20] and stimulated lipid peroxidation[21], with a possibly deleterious effect on the cardiovascular system as higher doses were administered. The daily vitamin C supplementation amount used was higher than the recommended daily intake for adults (90 mg/day for men and 75 mg/day for women), but lower than the maximum tolerable level (2000 mg/day)[22].

Reduced serum levels of α-tocopherol were inversely associated with the incidence of coronary artery disease[15], according to prospective observational studies in individuals without DM and/or without previous cardiovascular disease[23,24]. The α-tocopherol form of vitamin E is the most biologically active and could be considered a good biomarker of the consumption of this vitamin[25]. However, the beneficial effect observed in the study included in the current review occurred among users of antioxidant supplements, without specifying the supplement type and quantity[15]. Moreover, increased mortality from all causes[3] in subjects without DM was demonstrated with 10–5000 IU/day of vitamin E supplementation in randomized clinical trials. A possible explanation of the adverse effects described is that vitamin E can inhibit platelet function[26].

High serum zinc was shown to be protective against the development of cardiovascular disease[16], a result that is in accordance with other studies in patients without DM[27,28]. Patients with type 2 DM presented lower values of serum zinc (9.23 μmol/L *vs.* 12.46 μmol/L, $p < 0.001$) compared with patients without DM, suggesting a lower antioxidant capacity in diabetes[29]. Possibly, the importance of maintaining high serum zinc values is due to their role in an endogenous antioxidant system[30] and/or because zinc plays a clear role in the synthesis, storage and secretion of insulin[31].

Chromium[12] and selenium[14], which were measured in the toenail, were not associated with cardiovascular outcomes in patients with DM in the studies included in this review. This result is different from what was observed in subjects without DM in a case-control study performed in eight European countries and Israel (EURAMIC study)[32]. In the EURAMIC study, chromium levels in nails was inversely associated with the occurrence of myocardial infarction (OR 0.59; 95%CI 0.37–0.95)[32]. Moreover,

chromium[33] and zinc[31] are beneficial in regulating insulin action and energy metabolism. Better glycemic control could be reflected in lower cardiovascular outcomes[34]. In a recent systematic review with meta-analysis, chromium supplementation (1.28 to 1000 mcg/day) decreased the glycated hemoglobin values in 381 patients with DM by 0.6% (95%CI −0.9 to −0.2)[35]. However, cardiovascular outcomes were not evaluated in that study.

The study which evaluated selenium included in the current review was not in accordance with a recent meta-analysis of 25 observational studies[36] that demonstrated a reduction of 24% (95%CI 7–38) in the risk of coronary artery disease with an increase of 50% in selenium levels (assessed by different methods). Selenium is another essential mineral involved in antioxidant defense, since it is part of *glutathione peroxidase*, a selenoprotein. In this context, low serum selenium has been linked to increased risk of cardiovascular disease in subjects without DM[37]. The effects of selenium supplementation (200 μg/day) in the prevention of cardiovascular events were not confirmed in a randomized clinical trial[36] or in a prospective study having a follow-up of 7.6 years[38], probably due to its narrow therapeutic range. Selenium deficiency in humans appears to be just one factor in a complex set of nutritional variables that may predispose or protect against cardiovascular disease[37]. One of the limitations common to studies with selenium and chromium included in this review involves the measurement method adopted. Although the levels of these minerals in the toenail may reflect the long-term intake of the mineral[25], samples contamination could be a source of error[12,14].

Our systematic review has several limitations: 1. the low quality of the original studies; 2. no study included meets all items previously established to evaluate methodological quality and potential confounding factors were considered in the analysis of data just from four studies[12-14,16]; 3. no sensibility analysis was carried out due to clinical heterogeneity of studies included; 4. the case-control studies did not allow us to establish a cause-consequence between micronutrients intake and cardiovascular outcomes. Also, the results for supplementation of vitamin C derived from a single cohort study and need to be considered with caution.In conclusion and according to available evidence, information about antioxidant micronutrient intake and cardiovascular risk in individuals with DM is too scarce to determine which micronutrient antioxidants might be related to cardiovascular outcomes in the DM population. Moreover, the antioxidant property of micronutrients appears to be only one factor in a complex set of nutritional variables that may predispose or protect against cardiovascular disease. Further studies should be performed to explore the relationship between antioxidant micronutrient intake and the development of cardiovascular disease in patients with DM, preferably randomized controlled trials. The description of the results of this review will aid researchers interested in investigating the topic to develop their hypotheses.

Author contributions

Conception and design of the research, Acquisition of data and Critical revision of the manuscript for intellectual content: Sarmento RA, Silva FM, Sbruzzi G, Schaan BD, Almeida JC; Analysis and interpretation of the data: Sarmento RA, Silva FM, Schaan BD, Almeida JC; Statistical analysis: Sarmento RA, Silva FM, Sbruzzi G, Schaan BD; Writing of the manuscript: Sarmento RA.

Potential Conflict of Interest

No potential conflict of interest relevant to this article was reported.

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