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Review

# Can chocolate consumption reduce cardio-cerebrovascular risk? A systematic review and meta-analysis

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ABSTRACT

A systematic review and meta-analysis of the literature was performed to assess the relationship between chocolate intake and cardio-cerebrovascular risk in the general population. A structured search of the literature was performed in the PubMed database up to September 26, 2016, using predetermined keywords. Epidemiologic studies evaluating the risk for cardiovascular diseases (CVDs; i.e., stroke, acute myocardial infarction [MI], heart failure, coronary heart disease) were included according to different rates of chocolate intake. The software ProMeta 3 was used to perform the meta-analysis. The systematic review identified 16 eligible studies. The majority of the studies showed a protective effect of chocolate intake compared with unexposed individuals. The overall risk ratio (effect size [ES]) of CVD for the highest versus the lowest category of chocolate consumption was 0.77 (95% confidence interval [CI], 0.71–0.84; $P = 0.000$) with a moderate heterogeneity. The risk related to subgroups of CVD and in particular, the risk for MI was further analyzed: ES = 0.78 (95% CI, 0.64–0.94; $P = 0.009$) without statistical heterogeneity ($I^2 = 46.56\%$; $P = 0.13$). Moreover, the analysis performed based on sex found an ES = 0.85 (95% CI, 0.77–0.95; $P = 0.003$) for women, with a very low grade of heterogeneity ($I^2 = 62.21\%$; $P = 0.005$). The results of the meta-analysis showed a potential protective effect of moderate consumption of chocolate on cardiovascular risk, especially for women, and against MI for both sexes.



## Introduction

According to the latest epidemiologic update, cardiovascular disease (CVD) is the most common cause of death in Europe (45% of all deaths) [1]. CVD is a heterogeneous and complex group of diseases, including cerebrovascular disease (stroke), myocardial infarction (MI), and coronary heart disease (CHD). The mortality rate is higher for women (49%) than for men (40%) for both stroke and CHD [2,3]. A very high proportion (90%) of CVD is considered preventable [4] through a modification of lifestyle (moderate alcohol consumption, no smoking, physical activity, and healthy diet), blood pressure, and blood sugar control [5]. Some studies suggest that chocolate consumption might be inversely associated with prevalent calcified atherosclerotic plaques in the coronary arteries—a risk factor for CHD—as well as CHD incidence and mortality [6]. Chocolate is one of the most important dietary sources of flavonoids, polyphenolic compounds that may have cardioprotective effects due to hypothetical endothelial and platelet function [7] and important antioxidant action. Previous studies have shown that dietary intake of different types of flavonoids is associated with reduced risk for death from CHD and CVD [8]. Moreover, many flavonoids are potent antioxidants for low-density lipoprotein (LDL) oxidation, which is involved in the development of atherosclerotic diseases [9].

According to mythology, cocoa originated from the blood of an Aztec princess who preferred death rather than reveal the riches of her kingdom [10]. Mayas, Incas, and Aztecs cultivated

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the cocoa tree (*Theobroma cacao*) and praised it as "a gift of the gods." Chocolate is produced from the seeds of the cocoa tree [11]. In ancient history, chocolate was seen an aphrodisiac accessible only to the affluent and the rich [12] and numerous positive properties to human health have been ascribed to it. In the 18th century, without scientific evidence, cocoa was believed to strengthen the heart and reduce angina pectoris [13].

Cocoa beans are now known to contain very high levels of flavonols that occur both as monomers of epicatechin and catechin and as polymerized flavonols, or procyanidins. These substances are responsible for many protective effects: They can reduce platelet aggregation and modulate redox-mediated vasodilation as well as the transcription of inflammatory cytokines [14].

Although many studies observe a statistically significant [13] or a nonsignificant [15] inverse association between chocolate consumption and total stroke, a moderate consumption of chocolate might be associated with a lower risk for heart failure (HF). This benefit may be due to favorable effects of cocoa products on blood pressure, which is a major risk factor for HF [16].

In patients free of diabetes surviving their first acute myocardial infarction (AMI), moderate chocolate consumption is associated with lower cardiac mortality, suggesting that individuals with CHD do not need to avoid chocolate [15].

The review by Corti et al. [7] focused on potential mechanisms involved in the response to cocoa and the potential clinical implications associated with its consumption. The beneficial effects of cocoa are most likely due to an increased bioavailability of nitric oxide (NO), which has an effect on the endothelium, such as vasodilation and prevention of leukocyte adhesion and migration. Reduced NO bioavailability is associated with endothelial dysfunction and eventually atherosclerotic disease [7]. The aim of this meta-analysis was to evaluate the association between chocolate intake and risk for CVD in the general population. In particular, we also assessed the risk of specific subgroups of CVD such as HF, stroke, AMI, and CHD.

## Methods

A systematic review and meta-analysis in accordance with PRISMA guidelines was performed. The literature search was carried out on September 26, 2016, referring to PubMed, using predetermined keywords and a combination of Mesh terms, Title/Abstract, and text word. The search terms were selected based on three aspects: food intake, CVDs, and type of study; and finally, they were combined with Boolean operator AND/OR. The search terms related to the main aspects considered were:

- Food intake: Cacao*, cocoa*, chocolate*, "Diet/statistics and numerical data"[MeSH Terms];
- Cardiovascular disease: "Cardiovascular Diseases/epidemiology"[MAJR], "Myocardial Infarction/epidemiology"[MAJR], "Stroke/epidemiology"[MAJR], "Heart Failure/epidemiology"[MAJR], "Heart Failure/prevention and control"[MeSH Terms], coronary artery, atherosclerosis, ischemic heart, ischemic, ischemia, ischemia, cerebral stroke, brain vascular accident, cerebrovascular, cerebral vascular, CVA, CVD;
- Type of study: "Prospective Studies"[MeSH Terms], "Cross-Sectional Studies"[MeSH Terms], "Follow-Up Studies"[MeSH Terms], "Surveys and Questionnaires"[MeSH Terms], "Incidence"[MeSH Terms], "Cohort Studies"[MeSH Terms], epidemiology.

### *Inclusion criteria*

To be included in the current meta-analysis, the studies had be in English only, full length, performed on humans (no in vitro or animal studies), and focused on chocolate or cocoa intake, epidemiologic (case–control, cohort, cross-sectional studies) evaluating the relationship between chocolate/cocoa intake and risk for CVD (CVD, MI, stroke, ischemic heart disease, HF).

Exclusion criteria included different outcome studies without proper extraction data (e.g., odds ratio [OR], risk ratio [RR], hazard ratio [HR]), experimental animal models, and studies without original data (abstract, letters, comments, review).

To verify whether the retrieved studies satisfied the inclusion criteria, two reviewers (T.S. and V.G.) independently screened the titles and abstracts. Possible disagreements were resolved through discussion or third reviewer consultation. Full-text articles were downloaded for the selected titles. The reference lists of the retrieved articles were checked to identify additional publications.

### *Data extraction*

Two independent reviewers used a predefined spreadsheet to collect the main information from the included studies (T.S. and V.G.). The collected data included qualitative information of the studies (name of the first author, year of publication, type of study, country); participant characteristics (sample size, age range or mean age, sex, ethnicity, health status); characteristics of study design (recruitment procedures, duration of the enrolment, degree of exposure), instrument used to assess food intake, such as food frequency questionnaire (FFQ; validated or not, FFQ self-administered or by interview), and information on the outcomes. The first or last authors of the original studies were contacted by e-mail to reduce the number of studies excluded due to difficulties in extraction data process from the articles.

### *Quality evaluation*

Quality of the included study was evaluated independently by two reviewers (VG and TS), using the scoring system created on the basis of the Meta-analysis of Observational Studies in Epidemiology group [17], the Quality Assessment Tool for Systematic Reviews of Observational Studies [18], and the Strengthening the Reporting of Observational Studies in Epidemiology [19] and modified by Buitrago-Lopez et al. [20]. The scoring sheet allowed a total score of 0 to 6 points (6 reflecting the highest quality). The system accepts a mark of 0 or 1 point for each variable 1) justification given for the cohort; 2) appropriate inclusion and exclusion criteria; 3) diagnosis of CVD, based not only on self-report; 4) validated tool to assess chocolate intake; 6) adjustments were made for age, sex, body mass index (BMI), and smoking status; and 7) any other adjustments (such as for physical activity, dietary factors).

### *Statistical analysis*

The meta-analysis was performed by ProMeta 3 software. Heterogeneity among studies was evaluated using the $I^2$ statistics. The effect size (ES) was estimated by RR reported with its 95% confidence interval (CI). The statistical heterogeneity among studies was assessed by the $\chi^2$ test and $I^2$ statistic (high heterogeneity if $I^2$ >60%; $P > 0.10$) [21]. $P < 0.05$ was considered statistically significant. To calculate the pooled effect, a random-effects model was applied according to the found heterogeneity (Egger's linear regression test). Lastly, the funnel plot was visually evaluated to assess possible publication bias.

As studies evaluated different concentrations of chocolate consumption in terms of range, the individuals exposed to the highest level versus those exposed to the lowest level were compared.

### *Subgroup analysis*

To test the validity of the results, the meta-analysis was run according to different outcomes (CVD, HF, and stroke) and sex.

## Results

We identified 396 potential articles and, after a preliminary screening of the title and abstract, 361 were excluded because they were reviews (n = 49), in a different language (n = 18), or not relevant (n = 294). Overall, 35 articles were eligible; however, 10 studies were further excluded because data were not available and another 9 because different outcomes were analyzed. The selection flowchart is shown in Figure 1. The detailed reasons for exclusion are presented in Table 1 [22–40]. Tables 2–6 show the characteristics of studies included in the meta-analysis organized according clinical outcomes. Five studies were conducted in United States [6,8,16,43,50], 1 in Australia [45], and 10 in Europe. Of the 10 European studies, 5 were from Sweden [15,47–49,51], 1 from Spain [41], 2 from Germany [13,46], 1 from the United

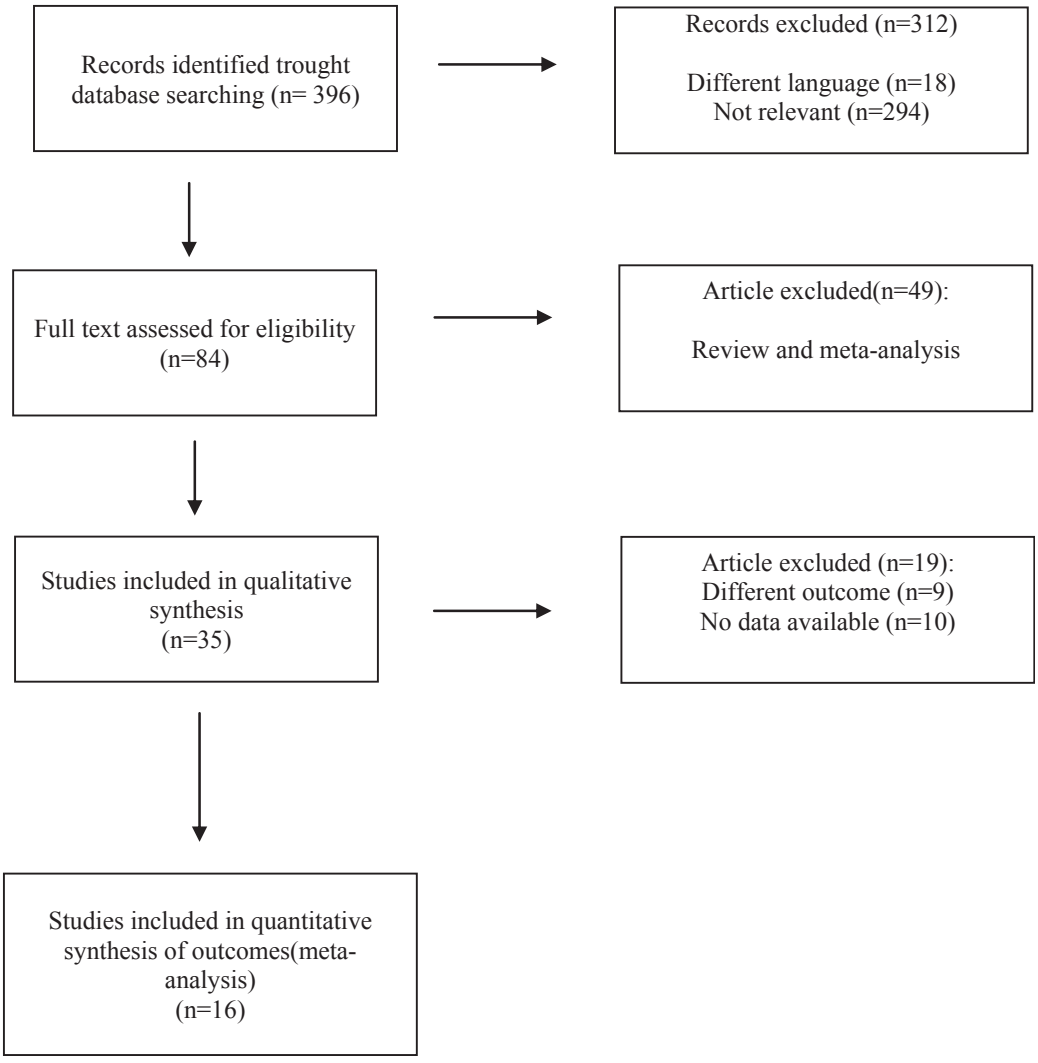


**Fig. 1.** Selection of studies flowchart, according to PRISMA's guidelines.

Kingdom [44], and 1 was a multicentric European study (Italy, Estonia, Cyprus, Belgium, Sweden, Germany, Hungary, and Spain) [42]. The results presented in the present meta-analysis are adjusted for at least four confounding factors such as BMI, sex, smoking habit, and age. Four studies evaluated the CVD risk in menopausal [51] and postmenopausal women [8,45,48,51], whereas four studies [16,43,49,46] evaluated the risk in adult men (age 45–79 y), and only one evaluated the risk for CVD in children ages 2 to 9 y [42]. The total risk for CVD was evaluated in five studies [7,13,15,44,42]. Myocardial infarction was evaluated by Lewis et al. [45], Larsson et al. [47–49], Buijsse et al. [13,46], and Janszky et al. [15]. Janszky et al. [15] also evaluated stroke, as did five other studies [8,13,48,49,44]. Lewis et al. evaluated carotid atherosclerotic plaques and atherosclerotic vascular disease [45]. Khawaja et al. estimated the risk for atrial fibrillation [43]; Alonso et al. evaluated the risk for hypertension [41]. CHD was investigated in four studies [6,8,50,44], whereas the risk for HF was observed in four other studies [15,16,45,51]. Because some of the included studies assessed more than one subgroup of CVD, results were considered as independent studies. Chocolate intake was estimated by self-administered FFQ previously validated in the majority of the studies; diversely, in Bel-Serrat et al. [42], the questionnaire was administered to the parents, one study used a cross-check history method adapted to the Dutch population [13], and in two studies [6,50], the questionnaire was administered by interview. Almost all were prospective studies, whereas three were cross-sectional [6,50,42]. The pooled sample consisted of 344 453 participants, and the ES was 0.71 (95% CI, 0.65–0.78; $P = 0.000$; Fig. 2A). The results of the present meta-analysis demonstrated an important effect in the highest versus lowest category of chocolate intake, although Egger's linear regression test shows a potential publication bias (intercept –2.04, $t = -2.79$; $P = 0.009$), also confirmed by the asymmetry of the Funnel plot (Fig. 2B). Moreover, high statistical heterogeneity ($\chi^2 = 182.49$; df = 32; $I^2 = 82.47\%$; $P = 0.000$) was found. To reduce heterogeneity, a further supplementary analysis excluding cross-sectional studies was conducted [6,50,42]. Although the ES was still significant at 0.77 (95% CI, 0.71–0.84; $P = 0.000$), heterogeneity was reduced ($\chi^2 = 103.81$; df = 29; $I^2 = 75.92\%$; $P = 0.000$), as was publication bias (Egger's linear regression test intercept –1.11, $t = -1.32$; $P = 0.198$; data not shown).

**Table 1**
Excluded studies and the reason for exclusion

| Author, year published [Ref no.] | Reason for exclusion |
|---|---|
| Arts et al. 2001 [22] | No data available |
| Bayard et al. 2007 [23] | |
| Brown et al. 2010 [24] | |
| Brummer 1969 [25] | |
| Droste et al. 2014 [26] | |
| Fisher et al. 2003 [27] | |
| Ivey et al. 2015 [28] | |
| Lloyd-Williams et al. 2009 [29] | |
| Panagiotakos et al. 2009 [30] | |
| Ponzo et al. 2015 [31] | |
| Donfrancesco et al. 2008 [32] | Different outcome |
| Greenberg 2015 [33] | |
| Greenberg and Buijsse 2013 [34] | |
| Horn et al. 2014 [35] | |
| Monagas et al. 2009 [36] | |
| K Hollenberg 2006 [37] | |
| Rose et al. 2010 [38] | |
| Vázquez-Agell et al. 2013 [39] | |
| Walters et al. 2013 [40] | |

*Acute myocardial infarction*

Three trials were conducted during the first decade of 2000 [15,45,46] and one in 2014 [16] (Table 3). The selection ranged from 1169 to 31 823 participants followed for ~10 y. The age of the participants ranged between 43 and 85 y. Janszky et al. analyzed nondiabetic patients hospitalized with a confirmed first AMI, enrolled in the Stockholm Heart Epidemiology Program (SHEEP) study [15]. Buijsse et al. evaluated the risk in patients without of AMI or stroke, without antihypertensive therapy of the Postdam arm of the European Prospective Investigation into Cancer and Nutrition (EPIC) study [13,46]. Lewis et al. [45] analyzed elderly women from a 5-y randomized controlled trial of calcium supplementation, and Larsson et al. used data from two prospective cohorts of men and women from the COSM (Cohort of Swedish Men) [49] and SMC (Swedish Mammography Cohort) studies [48], respectively. The values of ES reported in the articles indicate a moderate MI protective effect of chocolate in two cases [45,47], whereas in two studies no association was observed [15,46]. The pooled ES was 0.78 (95% CI, 0.64–0.94; $P = 0.009$) based on 79 001 participants (Fig. 3A). Moreover, no statistical heterogeneity ($\chi = 5.61$; df = 3; $I^2 = 46.56\%$; $P = 0.13$) was found. The funnel plot (Fig. 3B) shows no potential publication bias, which was confirmed by Egger's linear regression test (intercept −1.68, $t = -1.41$; $P = 0.295$; data not shown).

*Stroke*

Three trials were conducted during the first decade of 2000 [8,15,46] and another three between 2011 and 2015 [48,49,44] (Table 4). The selection ranged from 1169 to 37 103 participants. The follow-up period of the studies ranged between 8 and 16 y and the age of the participants ranged between 35 and 85 y. Janszky et al. analyzed SHEEP study participants [15]; Buijsse et al. evaluated the risk in patients of the Postdam arm of EPIC study [13,46]; Larsson et al. analyzed women from SMC [48]; and Mink et al. evaluated the risk in postmenopausal women in the Iowa Women's Healthy Study (IWHS) [8]. Larsson et al. used data from COSM [49], whereas Kwok et al. conducted a study using data from the EPIC-Norfolk cohort [44]. Larsson et al. independently evaluated the risk for total stroke, hemorrhagic stroke, and cerebral infarction [48,49]. Mink et al. considered stroke mortality [8]. The ES reported in the articles indicates a possible protective effect of stroke in two cases [48,46], whereas in four studies, the association was not statistically significant [8,15,49,44]. The pooled ES was 0.73 (95% CI, 0.63–0.86; $P = 0.000$) based on 123 482 participants (Fig. 4A). No statistical heterogeneity was found ($\chi^2 = 12.20$, df = 5, $I^2 = 59.03\%$; $P = 0.32$). The funnel plot (Fig. 4B) showed no potential publication bias, which was confirmed by Egger's linear regression test (intercept −1.94, $t = -1.10$; $P = 0.335$; data not shown). A supplementary analysis, excluding the Mink study, was conducted because this study tested the risk for stroke mortality instead of incidence. The lower risk for stroke incidence associated with chocolate consumption was still significant (ES = 0.70; 95% CI, 0.58–0.85; $P = 0.00029$; data not shown). However, a low statistical heterogeneity ($I^2 = 61.23\%$, $P = 0.035$) was found (data not shown) probably due to a low number of included studies.

*Coronary heart disease*

Of the four studies evaluating CHD, one was conducted during the first decade of 2000 [8] and the other three between 2011 and 2015 [6,50,44] (Table 5). The selection ranged from 2217 to 34 489 participants. The duration of the observation ranged between 2 and 16 y. The age of the participants ranged between 25 and 93 y. Djoussé et al. evaluated the risk among the participants of National Heart, Lung, and Blood Institute Family Heart (NHLBI) study [6,50]. Because Djoussé et al. calculated the risk for CHD among NHLBI participants with different baseline characteristics (Tables 2–6), the results were considered as independent studies in the meta-analysis. Kwok et al. examined the EPIC-Norfolk cohort [44]; Mink et al. conducted the study using data from IWHS [8]. The values of ES reported in the articles indicated a moderate CHD protective effect of chocolate in three cases [6,50,44], whereas in one study no association was observed [8]. The pooled sample was 49 425 individuals. Although this meta-analysis shows a high protective effect of chocolate intake on CHD (ES = 0.53; 95% CI, 0.40–0.71; $P = 0.000$) and the results show high heterogeneity ($\chi^2 = 75.88$, df = 6, $I^2 = 92.01\%$; $P = 0.000$; Fig. 5A). The funnel plot (Fig. 5B) shows a potential publication bias. These data were confirmed by Egger's test (intercept −5.35, $t = -2.93$; $P = 0.003$; data not shown).

*Heart failure*

Three trials were conducted during the first decade of 2000, and one in 2016 (Table 6). The selection ranged from 1169 to 31 823 participants, and the follow-up period ranged between 3 and 9.5 y. The age of the participants ranged between 43 and 84 y. Janszky et al. analyzed patients enrolled in SHEEP study [15]; Lewis et al. analyzed elderly women [45]; Petrone et al. analyzed the risk in men enrolled in PHS (Physician's Health Study) [16]; and Mostofsky et al. assessed the risk in women without baseline diabetes or a history of HF or AMI from the SMC [51]. The values of ES reported in the articles indicated a statistically significant protective effect of chocolate in two cases [45,51], whereas no association was observed in the other two studies [15,16]. The pooled ES was estimated on 14 117 participants (ES = 0.83; 95% CI, 0.55–1.26; $P = 0.392$; Fig. 6A). Furthermore, a high statistical heterogeneity ($\chi^2 = 14.86$, df = 3, $I^2 = 79.81\%$; $P = 0.002$) was found. The funnel plot (Fig. 6B) does not show a potential publication bias. These data were confirmed

**Table 2**
Characteristics extracted from the included studies (CVD and hypertension) and quality score

| First author, year published | No. in analysis | Age (y) | Baseline characteristics | Study period | Study type | Instrument | Outcome | Chocolate intake (g) and frequency | OR, RR, or HR (95% CI) | *P* value | Country | QS/6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Alonso 2005 [41] | 5880 | Mean 35.8 | Healthy subjects | 1999–2002 | Prospective cohort | FFQ validated self-administered | Hypertension | Highest vs lowest quintile | OR 1.1 (0.7–1.8) | 0.25 | Spain | 6 |
| Bel-Serrat (a) 2013 [42] | 5548 | 2–9 | Male children | 2007–2008 | Cross-sectional | FFQ validated parent administered | CVD | Highest tertile vs lowest | OR 0.20 (0.07–0.56) | | Europe (Italy, Estonia, Cyprus, Belgium, Sweden, Germany, Hungary, Spain) | 5 |
| Bel-Serrat (b) 2013 [42] | 5548 | 2–9 | Female children | 2007–2008 | Cross-sectional | FFQ validated parent administered | CVD | Highest vs lowest tertile | OR 0.53 (0.23–1.20) | | Europe (Italy, Estonia, Cyprus, Belgium, Sweden, Germany, Hungary, Spain) | 5 |
| Buijsse 2006 [13] | 470 | 65–84 | Men participating in the Zutphen Elderly Study, free of CVDs, diabetes mellitus, and cancer | 1985–2000 | Prospective cohort | Cross-check dietary history method | CVD mortality | 2.25 g/d | RR 0.50 (0.32–0.78) | 0.002 | Germany | 5 |
| Janszky 2009 (a) [15] | 1169 | 65–84 | SHEEP study | 1992–1994 | Prospective cohort | Survey self-administered | CVD mortality | 50 g $\geq$ 2/wk | HR 0.34 (0.17–0.70) | 0.01 | Sweden | 5 |
| Janszky 2009 (e) [15] | 1169 | 65–84 | SHEEP study | Follow-up 8 | Prospective cohort | Survey self-administered | Nonfatal CVD | 50 g $\geq$ 2/wk | HR 0.82 (0.59–1.14) | 0.30 | Sweden | 5 |
| Khawaja 2015 [43] | 18 819 | Mean 66 $\pm$ 9.1 | Male physicians in the PHS | Mean follow up 9 $\pm$ 3 y | Prospective cohort | FFQ validated self-administered | Atrial fibrillation | 1 ounce ($\sim$28.4 g) $\geq$5/wk | HR 1.05 (0.89–1.25) | 0.25 for linear trend | US | 5 |
| Kwok (a) 2015 [44] | 20 951 | 59 $\pm$ 9 | EPIC-Norfolk cohort | 1993–1997 follow up 2007 | Prospective cohort | FFQ self-administered | CVD | 15.6 to 98.8/d | HR 0.82 (0.72–0.95) | 0.003 | UK | 6 |
| Kwok (b) 2015 [44] | 20 951 | 59 $\pm$ 9 | EPIC-Norfolk cohort | 1993–1997 follow up 2008 | Prospective cohort | FFQ self-administered | Fatal CVD | 15.6 to 98.8/d | HR 0.71 (0.56–0.89) | 0.008 | UK | 6 |
| Lewis (a) 2010 [45] | 1216 | Mean 75 $\pm$ 3 | Old women of 5-y RCT of calcium supplements | 1998 followed for 9.5 y | Prospective cohort | FFQ validated self-administered | Atherosclerotic vascular disease | 0.38 $\pm$ 0.56 servings/d > 1 portion/wk | HR 0.76 (0.60–0.97) | 0.03 | Australia | 5 |
| Lewis (d) 2010 [45] | 1216 | Mean 75 $\pm$ 3 | Old women of 5-y RCT of calcium supplements | 1998 followed for 9.5 y | Prospective cohort | FFQ validated self-administered | Carotid plaques | 0.38 $\pm$ 0.56 servings/d > 1 portion/wk | HR 0.77 (0.60–0.98) | 0.04 | Australia | 5 |
| Mink (b) 2007 [8] | 34 489 | 55–69 | Postmenopausal women in the IWHS | 1986–2002 | Prospective cohort | FFQ validated self-administered | CVD mortality | 1 $\times$ /wk | RR 0.92 (0.84–1.00) | 0.062 | US | 5 |

The parenthetical letter indicate multiple used of the same source.
CI, confidence interval; CVD, cardiovascular disease; EPIC, European Prospective Investigation into Cancer and Nutrition; FFQ, food frequency questionnaire; HR, hazard ratio; IWHS, Iowa Women's Healthy Study; OR, odds ratio; PHS, Physicians Health Survey; QS/6, quality score; RCT, randomized controlled trial; RR, risk ratio; SHEEP, Stockholm Heart Epidemiology Program.

**Table 3**
Characteristics extracted from the included studies (outcome AMI) and quality score

| First author, year published | No in analysis | Age (y) | Baseline characteristics | Study period | Study type | Instrument | Outcome | Chocolate intake (g) and frequency | OR, RR, or HR (95% CI) | *P* value | Country | QS/6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Buijsse 2010 (a) [46] | 19 357 | 35–65 | Postdam arm of EPIC study | Mean follow-up 8.1 y | Prospective cohort | FFQ validated self-administered | AMI | Mean chocolate intake 6 g/d higher in the top quartile | RR 0.73 (0.47–1.15) | 0.33 linear trend | Germany | 5 |
| Janszky 2009 (b) [15] | 1169 | 65–84 | SHEEP study | Follow up 8 | Prospective cohort | Survey self-administered | Recurrent AMI | 50 g $\geq$ 2/wk | HR 0.86 (0.54–1.37) | 0.38 | Sweden | 5 |
| Larsson 2016 [47] | 67 640 | 45–83 | COSM and SMC | 1998–2010 | Prospective cohort | FFQ validated self-administered | AMI | $\geq$3 to 4 servings/wk | RR 0.87 (0.77–0.98) | 0.04 | Sweden | 5 |
| Lewis (b) 2010 [45] | 1216 | Mean 75 $\pm$ 3 | Old women of 5-y RCT of calcium supplements | 1998 followed for 9.5 y | Prospective cohort | FFQ validated self-administered | AMI | 0.38 $\pm$ 0.56 servings/d $>$ 1 portion/wk | HR 0.65 (0.46–0.94) | 0.02 | Australia | 5 |

The parenthetical letter indicate multiple used of the same source.
AMI, acute myocardial infarction; CI, confidence interval; COSM, Cohort of Swedish Men; EPIC, European Prospective Investigation into Cancer and Nutrition; FFQ, food frequency questionnaire; HR, hazard ratio; OR, odds ratio; QS/6, quality score; RCT, randomized controlled trial; RR, risk ratio; SHEEP, Stockholm Heart Epidemiology Program; SMC, Swedish Mammography Cohort.

**Table 4**
Characteristics extracted from the included studies (outcome stroke) and quality score

| First author, year published | No. in analysis | Age (y) | Baseline characteristics | Study period | Study type | Instrument | Outcome | Chocolate intake (g) and frequency | OR, RR, or HR (95% CI) | *P* value | Country | QS/6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Buijsse 2010 (b) [46] | 19 357 | 35–65 | Postdam arm of EPIC study | Mean follow-up 8.1 y | Prospective cohort | FFQ validated self-administered | Stroke | Mean chocolate intake 6 g/d higher in the top quartile | RR 0.52 (0.30–0.89) | 0.90 linear trend | Germany | 5 |
| Janszky 2009 (d) [15] | 1169 | 65–84 | SHEEP study | Follow up 8 | Prospective cohort | Survey self-administered | Stroke | 50 g $\geq$ 2/wk | HR 0.62 (0.33–1.16) | 0.65 | Sweden | 5 |
| Kwok (d) 2015 [44] | 20 951 | 59 $\pm$ 9 | EPIC-Norfolk cohort | 1993–1997 follow up 2010 | Prospective cohort | FFQ self-administered | Stroke | 15.6–98.8/d | HR 0.81 (0.62–1.05) | 0.14 | UK | 6 |
| Larsson (a) 2011 [48] | 33 372 | 49–83 | Women in SMC | 1997 followed until 2008 | Prospective cohort | FFQ validated self-administered | Total stroke | >45 g/wk | RR 0.80 (0.66–0.99) | 0.01 | Sweden | 6 |
| Larsson (b) 2011 [48] | 33 372 | 49–83 | Women in SMC | 1998 followed until 2008 | Prospective cohort | FFQ validated self-administered | Stroke | >45 g/wk | RR 0.83 (0.66–1.04) | 0.04 | Sweden | 6 |
| Larsson (c) 2011 [48] | 33 372 | 49–83 | Women in SMC | 1998 followed until 2008 | Prospective cohort | FFQ validated self-administered | Hemorrhagic stroke | >45 g/wk | RR 0.58 (0.34–1.00) | 0.04 | Sweden | 6 |
| Larsson (a) 2012 [49] | 37 103 | 45–79 | COSM | 1998–2008 | Prospective cohort | FFQ self-administered | Total stroke | 62.9 g/wk | RR 0.83 (0.70–0.99) | 0.08 | Sweden | 4 |
| Larsson (b) 2012 [49] | 37 103 | 45–79 | COSM | 1998–2008 | Prospective cohort | FFQ self-administered | Stroke | 62.9 g/wk | RR 0.83 (0.69–1.01) | 0.14 | Sweden | 4 |
| Larsson (c) 2012 [49] | 37 103 | 45–79 | COSM | 1998–2008 | Prospective cohort | FFQ self-administered | Hemorrhagic stroke | 62.9 g/wk | RR 0.84 (0.56–1.25) | 0.42 | Sweden | 4 |
| Mink (a) 2007 [8] | 34 489 | 55–69 | IWHS | 1986–2002 | Prospective cohort | FFQ validated self-administered | Stroke mortality | 1$\times$/wk | RR 0.85 (0.70–1.03) | 0.098 | US | 5 |

The parenthetical letter indicate multiple used of the same source.
CI, confidence interval; COSM, Cohort of Swedish Men; EPIC, European Prospective Investigation into Cancer and Nutrition; FFQ, food frequency questionnaire; HR, hazard ratio; IWHS, Iowa Women's Healthy Study; OR, odds ratio; QS/6, quality score; RR, risk ratio; SD, standard deviation; SHEEP, Stockholm Heart Epidemiology Program; SMC, Swedish Mammography Cohort.

**Table 5**
Characteristics extracted from the included studies (outcome CHD) and quality score

| First author, year published | No. in analysis | Age (y) | Baseline characteristics | Study period | Study type | Instrument | Outcome | Chocolate intake (g) and frequency | OR, RR, or HR (95% CI) | *P* value | Country | QS/6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Djoussé 2011 [6] | 2217 | 25.6–85.7 | NHLBI | 1993–2003 | Cross-sectional | FFQ validated staff-administered | CAC | 1 ounce (~28.4 g) ≥ 2/wk | OR 0.69 (0.48–0.99) | 0.029 for linear trend | US | 5 |
| Djoussé 2011 (a) [50] | 4970 | 25–93, mean 52 (SD 1.7) | NHLBI | 1993–1995 | Cross-sectional | FFQ validated staff-administered | CHD | 1 ounce > 5/wk | OR 0.43 (0.27–0.68) | 0.0002 | US | 5 |
| Djoussé 2011 (b) [50] | 4366 | 25–93, mean 52 (SD 13.7) | NHLBI | 1993–1995 | Cross-sectional | FFQ validated staff-administered | CHD | 1 ounce > 5/wk | OR 0.38 (0.23–0.63) | 0.0002 | US | 5 |
| Djoussé 2011 (c) [50] | 4790 | ≤60 | NHLBI | 1993–1995 | Cross-sectional | FFQ validated staff-administered | CHD | 1 ounce > 5/wk | OR 0.36 (0.17–0.75) | 0.0004 | US | 5 |
| Djoussé 2011 (d) [50] | 4790 | >60 | NHLBI | 1993–1995 | Cross-sectional | FFQ validated staff-administered | CHD | 1 ounce > 5/wk | OR 0.48 (0.28–0.83) | 0.016 | US | 5 |
| Kwok (c) 2015 [44] | 20 951 | 59 ± 9 | EPIC-Norfolk cohort | 1993–1997 follow up 2009 | Prospective cohort | FFQ self-administered | CHD | 15.6–98.8/d | HR 0.83 (0.71–0.97) | 0.006 | UK | 6 |
| Mink (c) 2007 [8] | 34 489 | 55–69 | IWHS | 1986–2002 | Prospective cohort | FFQ validated self-administered | CHD mortality | 1×/wk | RR 0.98 (0.88–1.10) | 0.775 | US | 5 |

The parenthetical letter indicate multiple used of the same source.
CAC, calcified atherosclerotic plaques in the coronary arteries; CHD, coronary heart disease; CI, confidence interval; CVD, cardiovascular disease; EPIC, European Prospective Investigation into Cancer and Nutrition; HR, hazard ratio; IWHS, Iowa Women's Healthy Study; NHLBI, National Heart, Lung, and Blood Institute; OR, odds ratio; QS/6, quality score; RR, risk ratio; SD, standard deviation.

**Table 6**
Characteristics extracted from the included studies (outcome heart failure) and quality score

| First author, year published | No. in analysis | Age (y) | Baseline characteristics | Study period | Study type | Instrument | Outcome | Chocolate intake (g) and frequency | OR, RR, or HR (95% CI) | *P* value | Country | QS/6 |
|---|---|---|---|---|---|---|---|---|---|---|---|---|
| Janszky 2009 (c) [15] | 1169 | 65–84 | SHEEP study | Follow up 8 y | Prospective cohort | Survey self-administered | Congestive HF | 50 g ≥ 2/wk | HR 0.78 (0.52–1.16) | 0.78 | Sweden | 5 |
| Lewis (c) 2010 [45] | 1216 | Mean 75 ± 3 | Old women of 5-y RCT of calcium supplements | 1998 followed for 9.5 y | Prospective cohort | FFQ validated self-administered | HF events | 0.38 ± 0.56 servings/d > 1 portion/wk | HR 0.41 (0.22–0.76) | 0.01 | Australia | 5 |
| Mostofsky 2010 [51] | 31 823 | 43–83 | Women in SMC | 1998–2006 | Prospective cohort | FFQ self-administered | HF | ≥1 servings/d | Rate ratios 1.23 (0.73–2.08) | 0.0005 for quadratic trend | Sweden | 5 |
| Petrone 2014 [16] | 20 278 | 66.4 ± 9.2 | Men from PHS | 1999–2002 | Prospective cohort | FFQ validated self-administered | HF | 1 ounce (~28.4 g) > 5/wk | HR 0.82 (0.63–1.07) | 0.41 for linear trend, 0.62 for quadratic trend | US | 4 |

The parenthetical letter indicate multiple used of the same source.
CI, confidence interval; FFQ, food frequency questionnaire; HF, heart failure; HR, hazard ratio; NHLBI, National Heart, Lung, and Blood Institute; OR, odds ratio; PHS, Physicians Health Survey; QS/6, quality score; RCT, randomized controlled trial; RR, risk ratio; SHEEP, Stockholm Heart Epidemiology Program; SMC, Swedish Mammography Cohort.

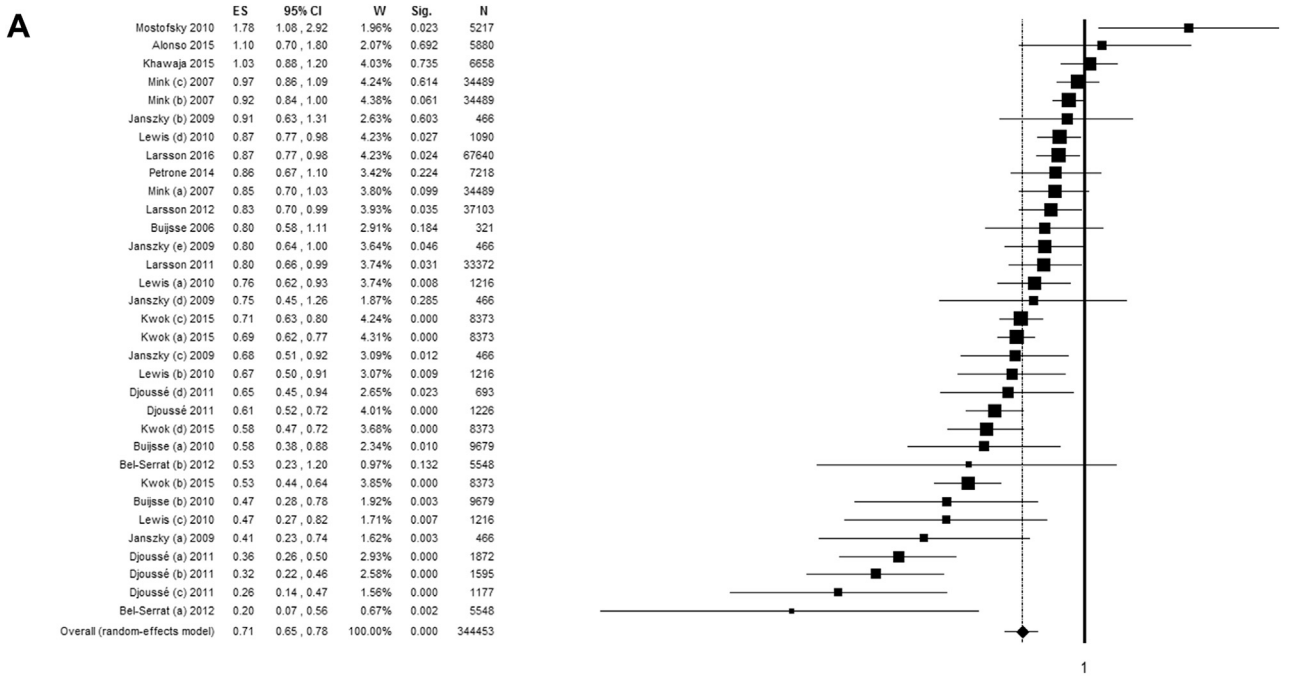


**Fig. 2.** (A) Forest and (B) funnel plots of the meta-analysis comparing chocolate intake in the prevention of CVDs (16 epidemiological studies). CVD, cardiovascular disease; ES, effect size.

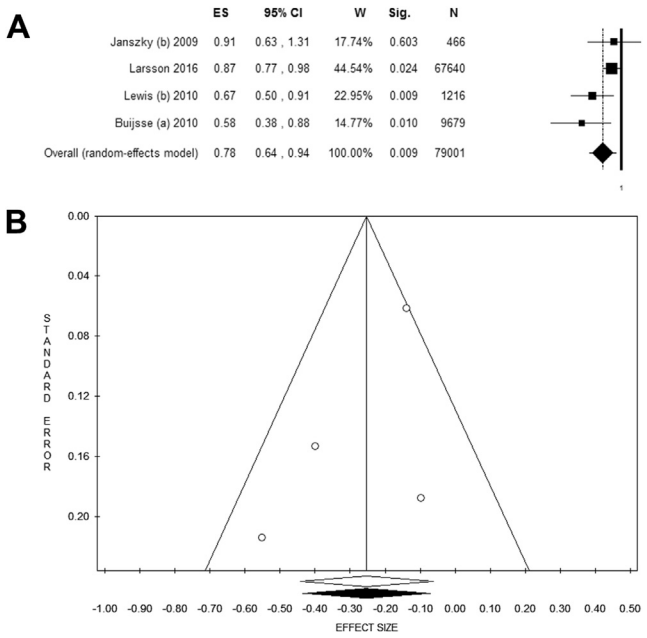


**Fig. 3.** (A) Forest and (B) funnel plots of the meta-analysis comparing chocolate intake in the prevention of AMIs (4 epidemiological studies). AMI, acute myocardial infarction; ES, effect size.

by Egger's test (intercept 0.61, $t = 0.14$; $P = 0.901$; data not shown).

*Cardiovascular risk by sex*

For a more comprehensive evaluation, the meta-analysis was performed according to sex. There were five articles that appraised the risk for CVD in women [8,45,48,51,42], but because some of them assessed the risk for different CVDs such as atherosclerotic vascular disease, HF, and carotid plaques, these were considered as separate studies. All the studies evaluated the risk in menopausal or postmenopausal women (ages 43–83 y), except the study by Bel-Serrat et al. [42], which estimated the risk in female children ages 2 to 9 y. The total sample size was 152 342 and the ES was statistically significant at 0.85 (95% CI; 0.77–0.95; $P = 0.003$) with a low grade of heterogeneity ($\chi^2 = 23.81$, df = 9, $I^2 = 62.21\%$; $P = 0.005$; Fig. 7A). The funnel plot shows a low-grade potential publication bias, confirmed by Egger's test (intercept –1.21, $t = 1.27$; $P = 0.239$; data not shown).

The same analysis was performed for men, although there were four studies included [16,43,49,42]. Three of these studies were conducted in men; mean age was ~60 y and the outcomes were atrial fibrillation, HF, and stroke. The fourth study was conducted in male children ages 2 to 9 y, in eight European countries (Italy, Estonia, Cyprus, Belgium, Sweden, Germany, Hungary, Spain). The pooled ES was evaluated on a total sample of 56 527 participants. The results show a nonstatistically significant protective effect (ES = 0.84; 95% CI, 0.66–1.07; $P = 0.164$) with a moderate grade of heterogeneity ($\chi^2 = 11.74$, df = 3, $I^2 = 74.44\%$; $P = 0.008$; Fig. 7B). The funnel plot shows no potential publication bias, confirmed by Egger's test (intercept –3.43, $t = -2.64$; $P = 0.118$; data not shown).

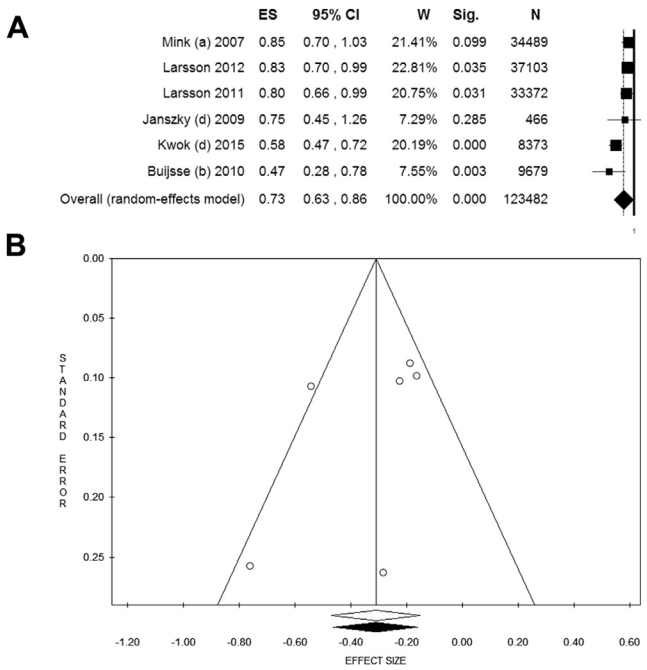


**Fig. 4.** (A) Forest and (B) funnel plots of the meta-analysis comparing chocolate intake in the prevention of stroke (6 epidemiological studies). ES, effect size.

## Discussion

The present meta-analysis showed that chocolate consumption was associated with a significant reduced risk for CVD (29%); in particular, 22% for AMI, 30% for stroke, 17% for HF, and 47% for CHD. Two studies further classified stroke as cerebral infarction and hemorrhagic stroke, but due to the low number of studies evaluating the differences between these two types of stroke, it was not possible to analyze the respective risk in this meta-analysis. Nevertheless, Larsson et al. [48,49] found in a multivariable model that the RR of cerebral infarction after an

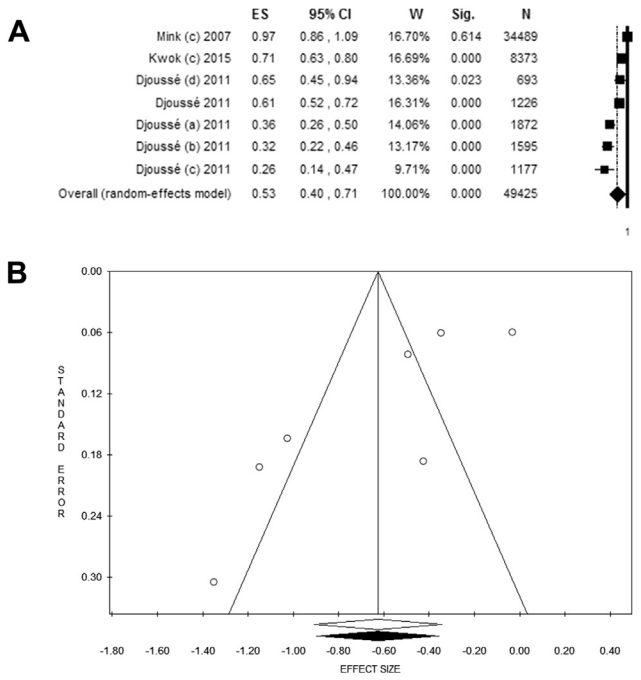


**Fig. 5.** (A) Forest and (B) funnel plots of the meta-analysis comparing chocolate intake in the prevention of CHDs (4 epidemiological studies). CHD, coronary heart disease; ES, effect size.

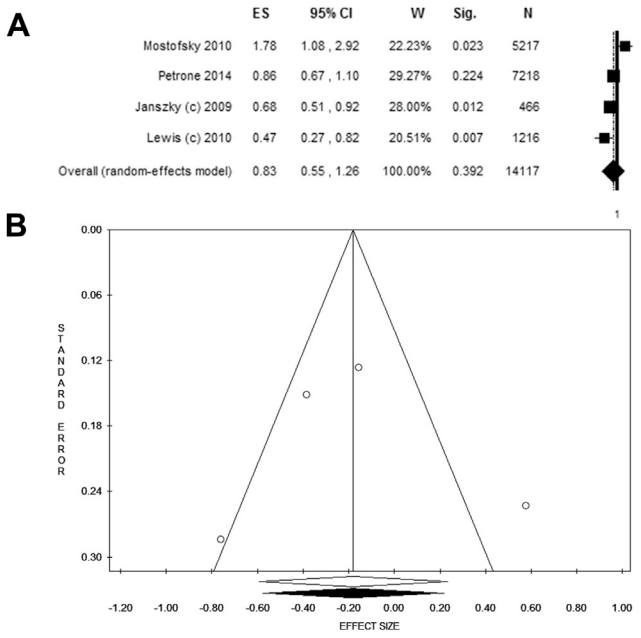


**Fig. 6.** (A) Forest and (B) funnel plots of the meta-analysis comparing chocolate intake in the prevention of HF (4 epidemiological studies). HF, heart failure; ES, effect size.

increased chocolate intake of 50 g/wk was 0.88 (95% CI, 0.54–0.99) in women and 0.83 (95% CI, 0.69–1.01) in men, whereas the RR for hemorrhagic stroke was 0.73 (95% CI, 0.54–0.99) in women and 0.84 (95% CI, 0.56–1.25) in men. Moreover, Djoussé et al. [50] also evaluated the association between chocolate consumption and risk for hypertension. Although they compared the lowest with the highest category of chocolate intake (>5 servings/wk), the adjusted odds ratio (OR) was 1.11 (95% CI, 0.81–1.53). This meta-analysis confirmed the results previously obtained in another meta-analysis where chocolate consumption was correlated to 37% reduction of CVD, 29% reduction of stroke, and 31% reduction of diabetes [20].

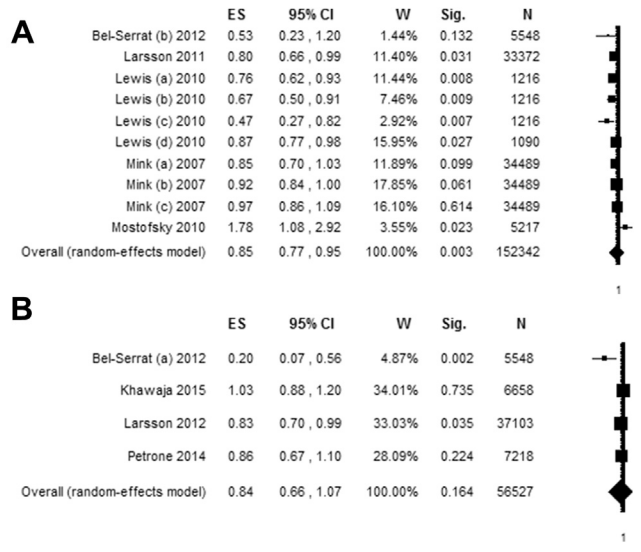


**Fig. 7.** Forest plots of the meta-analysis comparing chocolate intake in the prevention of cardiovascular risk by sex (A) women; (B) men. ES, effect size.

The majority of the included studies reported a significant reduction of CVD risk in association with higher levels of chocolate intake after adjustment for potential confounders, including age, physical activity, BMI, smoking status, dietary factor, education, and drug use.

Some issues need to be taken into account when interpreting the results of the present meta-analysis. Although FFQ is a well-established method for quantifying dietary information, it has some limitations: Chocolate intake could be underestimated because chocolate and cocoa are often used as ingredients in food formulation. In some cases, the FFQ could not distinguish between milk chocolate and dark chocolate. This distinction is important because the cocoa content is lower in milk than in dark chocolate; furthermore, milk could reduce the bioavailability of flavonols [52]. Another limitation is that chocolate intake was self-reported and measured at baseline only. Misclassification or underreporting could result in a low grade of association. Also, individuals might consume chocolate in a dietary pattern characterized by frequent snacking and high-energy foods. The high caloric value provided by high content of sugar and fat of commercial chocolate snack should be taken into account before generalizing the results. Bias due to different categories of chocolate intake established in the primary studies is also possible. In fact, it is possible that some of the studies have defined the cutoff differently in postanalysis (serving, portion, rare versus frequent, etc.). All the aforementioned limitations can explain the different results between trial and observational studies included in the present analysis. Moreover, the lack of clear information on the type of chocolate (milk, dark, or white) being consumed in primary studies did not allow us to identify which chocolate provides the biological and health-promoting effects. Probably, the protective effect should be ascribed to cocoa. Future studies need to focus on this particular aspect.

The strength of the present meta-analysis is the large number of participants (344 453), both women and men ranging from 2 to 93 y of age. Another strength of this study is the subgroup analysis. Actually, knowledge about intervention effectiveness in different subgroups is particularly important for a decision maker in public health [53]. To the best of our knowledge, this is the first meta-analysis to assess the risk for CVD separately in women and men. As previously mentioned, women have the highest mortality rate [1], and at the same time in the present meta-analysis, women represented the group that realizes the most important beneficial effects of chocolate consumption. Further epidemiologic studies are needed to confirm these results and to quantify the effects of behavior change in this particular category. Moreover, the majority of the included studies performed a complete follow-up (a mean of ~10 y). Another important aspect of the present analysis is that the risk for the different outcomes (AMI, stroke, HF, CHD) was considered simultaneously and separately. To obtain more accurate data and to minimize statistical errors, some of the study authors were contacted by e-mail.

## Conclusion

Findings from the present analysis agree on a potential beneficial association of chocolate consumption with a lower risk for CVDs. These results do not exclude that overconsumption of chocolate/cocoa can have harmful effects. Further studies are required to confirm these data before any recommendations about chocolate intake can be made. In particular, it is important to focus attention on the different types of chocolate (milk or dark chocolate, and chocolate or cocoa in snacks). In fact, snacks

with or without chocolate are very rich in sugar, saturated fat, and calories and for this reason they should be consumed in moderation. Moreover, the included studies do not specify the percentage of cocoa intake nor the type of chocolate consumed. This limit does not allow a clear indication of which chocolate is responsible for the beneficial effect, but it can probably be ascribed to the amount of cocoa in the chocolate. Future studies are needed on these aspects.

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## References

[1] Townsend N, Wilson L, Bhatnagar P, Wickramasinghe K, Rayner M, Nichols M. Cardiovascular disease in Europe: epidemiological update 2016. Eur Heart J 2016;37:3232–45.
[2] World Health Organization (WHO). Age-standardized death rates per 100,000 by cause. 2016. Available at: http://www.who.int/whosis/whostat2006 AgeStandardizedDeathRates.pdf?ua=1. Accessed February 10, 2017.
[3] Nichols M, Townsend N, Luengo-Fernandez R, Leal J, Gray A, Scarborough P, et al. European cardiovascular disease statistics 2012. Brussels, Belgium and Sophia Antipolis, France: European Heart Network and European Society of Cardiology; 2012.
[4] McGill HC Jr, McMahan CA, Gidding SS. Preventing heart disease in the 21st century: implications of the Pathobiological Determinants of Atherosclerosis in Youth (PDAY) study. Circulation 2008;117:1216–27.
[5] Finks SW, Airee A, Chow SL, Macaulay TE, Moranville MP, Rogers KC, et al. Key articles of dietary interventions that influence cardiovascular mortality. Pharmacotherapy 2012;32:e54–87.
[6] Djoussé L, Hopkins PN, Arnett DK, Pankow JS, Borecki I, North KE, et al. Chocolate consumption is inversely associated with calcified atherosclerotic plaque in the coronary arteries: the NHLBI Family Heart Study. Clin Nutr 2011;30:38–43.
[7] Corti R, Flammer AJ, Hollenberg NK, Luscher TF. Cocoa and cardiovascular health. Circulation 2009;119:1433–41.
[8] Mink PJ, Scrafford CG, Barraj LM, Harnack L, Hong CP, Nettleton JA, et al. Flavonoid intake and cardiovascular disease mortality: a prospective study in postmenopausal women. Am J Clin Nutr 2007;85:895–909.
[9] Waterhouse AL, Shirley JR, Donovan JL. Antioxidants in chocolate. Lancet 1996;348:834.
[10] Verna R. The history and science of chocolate. Malays J Pathol 2013;35:111–21.
[11] Bruinsma K, Taren DL. Chocolate: Food or drug? J Am Diet Assoc 1999;99:1249–56.
[12] Latif R. Chocolate/cocoa and human health: a review. Neth J Med 2013;71:63–8.
[13] Buijsse B, Feskens EJ, Kok FJ, Kromhout D. Cocoa intake, blood pressure, and cardiovascular mortality: the Zutphen Elderly Study. Arch Intern Med 2006;166:411–7.
[14] Scheid L, Reusch A, Stehle P, Ellinger S. Antioxidant effects of cocoa and cocoa products ex vivo and in vivo: is there evidence from controlled intervention studies? Curr Opin Clin Nutr Metab Care 2010;13:737–42.
[15] Janszky I, Mukamal KJ, Ljung R, Ahnve S, Ahlbom A, Hallqvist J. Chocolate consumption and mortality following a first acute myocardial infarction: the Stockholm Heart Epidemiology Program. J Intern Med 2009;266:248–57.
[16] Petrone AB, Gaziano JM, Djoussé L. Chocolate consumption and risk of heart failure in the Physicians' Health Study. Eur J Heart Fail 2014;16:1372–6.
[17] Stroup DF, Berlin JA, Morton SC, Olkin I, Williamson GD, Rennie D, et al. Meta-analysis of observational studies in epidemiology: a proposal for reporting. Meta-analysis Of Observational Studies in Epidemiology (MOOSE) group. JAMA 2000;283:2008–12.
[18] Wong WC, Cheung CS, Hart GJ. Development of a quality assessment tool for systematic reviews of observational studies (QATSO) of HIV prevalence in men having sex with men and associated risk behaviours. Emerg Themes Epidemiol 2008;5:23.
[19] von Elm E, Altman DG, Egger M, Pocock SJ, Gotzsche PC, Vandenbroucke JP, et al. The Strengthening the Reporting of Observational Studies in Epidemiology (STROBE) statement: guidelines for reporting observational studies. J Clin Epidemiol 2008;61:344–9.
[20] Buitrago-Lopez A, Sanderson J, Johnson L, Warnakula S, Wood A, Di Angelantonio E, et al. Chocolate consumption and cardiometabolic disorders: systematic review and meta-analysis. BMJ 2011;343:d4488.
[21] Higgins JP, Thompson SG, Deeks JJ, Altman DG. Measuring inconsistency in meta-analyses. BMJ 2003;327:557–60.
[22] Arts IC, Hollman PC, Feskens EJ, Bueno de Mesquita HB, Kromhout D. Catechin intake might explain the inverse relation between tea consumption and ischemic heart disease: the Zutphen Elderly Study. Am J Clin Nutr 2001;74:227–32.
[23] Bayard V, Chamorro F, Motta J, Hollenberg NK. Does flavanol intake influence mortality from nitric oxide-dependent processes? Ischemic heart disease, stroke, diabetes mellitus, and cancer in Panama. Int J Med Sci 2007;4:53–8.
[24] Brown SC, Geiselman PJ, Broussard T. Cardiovascular risk in African American women attending historically Black colleges and universities: the role of dietary patterns and food preferences. J Health Care Poor Underserved 2010;21:1184–93.
[25] Brummer P. Coronary mortality and living standard. II. Coffee, tea, cocoa, alcohol and tobacco. Acta Med Scand 1969;186:61–3.
[26] Droste DW, Iliescu C, Vaillant M, Gantenbein M, De Bremaeker N, Lieunard C, et al. Advice on lifestyle changes (diet, red wine and physical activity) does not affect internal carotid and middle cerebral artery blood flow velocity in patients with carotid arteriosclerosis in a randomized controlled trial. Cerebrovasc Dis 2014;37:368–75.
[27] Fisher ND, Hughes M, Gerhard-Herman M, Hollenberg NK. Flavanol-rich cocoa induces nitric-oxide-dependent vasodilation in healthy humans. J Hypertens 2003;21:2281–6.
[28] Ivey KL, Hodgson JM, Croft KD, Lewis JR, Prince RL. Flavonoid intake and all-cause mortality. Am J Clin Nutr 2015;101:1012–20.
[29] Lloyd-Williams F, Mwatsama M, Ireland R, Capewell S. Small changes in snacking behaviour: the potential impact on CVD mortality. Public Health Nutr 2009;1:871–6.
[30] Panagiotakos D, Pitsavos C, Chrysohoou C, Palliou K, Lentzas I, Skoumas I, et al. Dietary patterns and 5-year incidence of cardiovascular disease: a multivariate analysis of the ATTICA study. Nutr Metab Cardiovasc Dis 2009;19:253–63.
[31] Ponzo V, Goitre I, Fadda M, Gambino R, De Francesco A, Soldati L, et al. Dietary flavonoid intake and cardiovascular risk: a population-based cohort study. J Transl Med 2015;13:218.
[32] Donfrancesco C, Lo Noce C, Brignoli O, Riccardi G, Ciccarelli P, Dima F, et al. Italian network for obesity and cardiovascular disease surveillance: a pilot project. BMC Fam Pract 2008;9:53.
[33] Greenberg JA. Chocolate intake and diabetes risk. Clin Nutr 2015;34:129–33.
[34] Greenberg JA, Buijsse B. Habitual chocolate consumption may increase body weight in a dose-response manner. PLoS One 2013;8:e70271.
[35] Horn P, Amabile N, Angeli FS, Sansone R, Stegemann B, Kelm M, et al. Dietary flavanol intervention lowers the levels of endothelial microparticles in coronary artery disease patients. Br J Nutr 2014;111:1245–52.
[36] Monagas M, Khan N, Andres-Lacueva C, Casas R, Urpi-Sarda M, Llorach R, et al. Effect of cocoa powder on the modulation of inflammatory biomarkers in patients at high risk of cardiovascular disease. Am J Clin Nutr 2009;90:1144–50.
[37] Hollenberg K. N. Vascular action of cocoa flavanols in humans: the roots of the story. J Cardiovasc Pharmacol 2006;47(suppl 2):S99–102. discussion S19–21.
[38] Rose N, Koperski S, Golomb BA. Mood food: chocolate and depressive symptoms in a cross-sectional analysis. Arch Intern Med 2010;170:699–703.
[39] Vázquez-Agell M, Urpi-Sarda M, Sacanella E, Camino-Lopez S, Chiva-Blanch G, Llorente-Cortes V, et al. Cocoa consumption reduces NF-kappaB activation in peripheral blood mononuclear cells in humans. Nutr Metab Cardiovasc Dis 2013;23:257–63.
[40] Walters MR, Williamson C, Lunn K, Munteanu A. Chocolate consumption and risk of stroke: a prospective cohort of men and meta-analysis. Neurology 2013;80:1173–4.
[41] Alonso A, de la Fuente C, Beunza JJ, Sanchez-Villegas A, Martinez-Gonzalez MA. Chocolate consumption and incidence of hypertension. Hypertension 2005;46:e21–2. author reply e2.
[42] Bel-Serrat S, Mouratidou T, Bornhorst C, Peplies J, De Henauw S, Marild S, et al. Food consumption and cardiovascular risk factors in European children: the IDEFICS study. Pediatr Obes 2013;8:225–36.
[43] Khawaja O, Petrone AB, Kanjwal Y, Gaziano JM, Djoussé L. Chocolate consumption and risk of atrial fibrillation (from the Physicians' Health Study). Am J Cardiol 2015;116:563–6.
[44] Kwok CS, Boekholdt SM, Lentjes MA, Loke YK, Luben RN, Yeong JK, et al. Habitual chocolate consumption and risk of cardiovascular disease among healthy men and women. Heart 2015;101:1279–87.
[45] Lewis JR, Prince RL, Zhu K, Devine A, Thompson PL, Hodgson JM. Habitual chocolate intake and vascular disease: a prospective study of clinical outcomes in older women. Arch Intern Med 2010;170:1857–8.
[46] Buijsse B, Weikert C, Drogan D, Bergmann M, Boeing H. Chocolate consumption in relation to blood pressure and risk of cardiovascular disease in German adults. Eur Heart J 2010;31:1616–23.
[47] Larsson SC, Akesson A, Gigante B, Wolk A. Chocolate consumption and risk of myocardial infarction: a prospective study and meta-analysis. Heart 2016;102:1017–22.

[48] Larsson SC, Virtamo J, Wolk A. Chocolate consumption and risk of stroke in women. J Am Coll Cardiol 2011;58:1828–9.
[49] Larsson SC, Virtamo J, Wolk A. Chocolate consumption and risk of stroke: a prospective cohort of men and meta-analysis. Neurology 2012;79:1223–9.
[50] Djoussé L, Hopkins PN, North KE, Pankow JS, Arnett DK, Ellison RC. Chocolate consumption is inversely associated with prevalent coronary heart disease: the National Heart, Lung, and Blood Institute Family Heart Study. Clin Nutr 2011;30:182–7.
[51] Mostofsky E, Levitan EB, Wolk A, Mittleman MA. Chocolate intake and incidence of heart failure: a population-based prospective study of middle-aged and elderly women. Circ Heart Fail 2010;3:612–6.
[52] Mullen W, Borges G, Donovan JL, Edwards CA, Serafini M, Lean ME, et al. Milk decreases urinary excretion but not plasma pharmacokinetics of cocoa flavan-3-ol metabolites in humans. Am J Clin Nutr 2009;89:1784–91.
[53] Veroniki AA, Straus SE, Ashoor H, Stewart LA, Clarke M, Tricco AC. Contacting authors to retrieve individual patient data: study protocol for a randomized controlled trial. Trials 2016;17:138.