Clinical Nutrition ESPEN 42 (2021) 1–14

Contents lists available at ScienceDirect

Clinical Nutrition ESPEN

journal homepage: http://www.clinicalnutritionespen.com

Narrative Review

Effects of vitamin supplements on clinical cardiovascular outcomes: Time to move on! – A comprehensive review

Bahadir Simsek [a], Atakan Selte [a], Bugra Han Egeli [b], Ufuk Çakatay [c, *]

[a] Cerrahpasa Medical School, Istanbul University-Cerrahpasa, 34098, Fatih/Istanbul, Turkey
[b] Graduate Medical Sciences, Boston University School of Medicine, 72 E Concord St L-317, 02118, Boston, MA, USA
[c] Cerrahpasa Medical School, Department of Medical Biochemistry, Istanbul University-Cerrahpasa, 34098, Fatih/Istanbul, Turkey

ARTICLE INFO

Article history:
Received 25 November 2020
Accepted 5 February 2021

Keywords:
Cardiovascular health
Clinical outcomes
COVID-19
Dietary supplements
Vitamin D
Vitamin supplementation

SUMMARY

Background & aims: Vitamin supplementations have increasingly been advertised on media and reported to be widely used by the general public to improve cardiovascular health. Due to the COVID-19 pandemic, people have become more interested in ways to improve and maintain their health. Increased awareness of people on healthy lifestyle is translating into inquisition regarding dietary supplements. Aim: First, focus on the most commonly used vitamin supplements and comprehensively review the evidence for and against recommending them to patients to improve and/or maintain cardiovascular health. Second, illustrate how the interest in studies shifted over time from Vitamin A, E, C, and B to Vitamin D and observational studies led to randomized controlled trials.

Methods: A thorough PubMed search with the phrase: "Vitamin supplements and cardiovascular health" was performed. In the present review, focus was maintained on the evidence for the use of vitamin supplements in the prevention of major cardiovascular events and/or the maintenance of cardiovascular health by comprehensively reviewing all previous studies indexed in PubMed. Studies with clinical 'hard' end-points were included only.

Results: A total of 87 studies met the inclusion criteria and were reviewed in the present article. High-quality evidence suggesting benefits for the use of vitamin supplements to maintain or improve cardiovascular health in people is minimal to non-existent.

Conclusions: Vitamin supplementation does not improve clinical cardiovascular outcomes in general population. Counseling on the importance of maintaining a healthy lifestyle with adequate and nutritious food intake seems more appropriate to improve and maintain cardiovascular health.

© 2021 European Society for Clinical Nutrition and Metabolism. Published by Elsevier Ltd. All rights reserved.

1. Introduction

In recent years, better management of major cardiovascular events and preventive medicine have led to a decrease in cardiovascular mortality in developed countries [1]. Despite the increased evidence-based knowledge as to how to manage cardiovascular diseases (CVDs), CVDs continue to remain as a leading cause of mortality and morbidity in developed countries, and have been increasing globally [1]. Use of vitamin supplements has become even more popular in recent years with as many as 72% of older adults (age >65 years) in the US reporting consumption [2]. Increased interest regarding the maintenance of healthy lifestyle, absence of scientific understanding of laypeople regarding the utility of dietary supplements on the treatment and prevention of diseases combined with the look for a panacea further fostered interest in vitamin supplements in general population. Other than a few specific subgroups of population, such as to reduce the risk of malnutrition in disadvantaged populations, meet the demand of anabolism in pregnancy and childhood, and promote muscle build-up in body builders, the evidence supporting the benefits of regular vitamin supplementation is not proven [3]. The largest market for supplements is North America and the global market for dietary supplements is estimated to be around 123 billion USD and is expected to grow by 8.2%, reaching 230.73 billion USD by 2027. Most commonly used and sought for supplements include vitamins and minerals. In the US, 52% of adults reported using at least one

* Corresponding author.
E-mail addresses: bahadirsimsekk@gmail.com (B. Simsek), atakanselte@gmail.com (A. Selte), bhegeli@bu.edu (B.H. Egeli), ucakatay@gmail.com (U. Çakatay).

https://doi.org/10.1016/j.clnesp.2021.02.014
2405-4577/© 2021 European Society for Clinical Nutrition and Metabolism. Published by Elsevier Ltd. All rights reserved.

Abbreviations

ABI	Ankle-brachial index
AFib	Atrial fibrillation
BP	Blood pressure
CAD	Coronary artery disease
CCA-IMT	Common carotid artery intima-media thickness
CHD	Coronary heart disease
CKD	Chronic kidney disease
COVID-19	Coronavirus Disease 2019
CVD	Cardiovascular disease
DM	Diabetes mellitus
FMD	Flow mediated dilatation
IHD	Ischemic heart disease
IMT	Intima-media thickness
MI	Myocardial infarction
NEJM	New England Journal of Medicine
PAD	Peripheral artery disease
RR	Relative risk
RCT	Randomized clinical trial
PWV	Pulse wave velocity

supplement and vitamins were the leading with 48% [4]. In office visits during the COVID-19 era, physicians are increasingly being queried by their patients to provide advice on the use of nutritional supplements for reasons such as to strengthen their immune system and improve cardiovascular health. Therefore, the aim of the present review article is two-fold: First, to focus on the most commonly used vitamin supplements and comprehensively review the empirical evidence for and against recommending them to patients to improve and/or maintain cardiovascular health. Second, to illustrate how the interest in vitamin supplementation studies has shifted over time from Vitamin E, C, and B to Vitamin D and observational studies led to randomized controlled trials (RCTs).

2. Methods

2.1. Search strategy

The following phrase: "Vitamin supplements and cardiovascular health" was searched to date (October 2020) on PubMed. To prevent the possibility of missing important original research articles that were not found under the first round of PubMed search, the articles found in the first round (1384 articles) were also reviewed for relevance; by checking their title, study type (e.g., review article or original investigation), and reading their abstract, and in cases where relevant, they were also reviewed for inclusion. In cases where abstracts did not include enough information for inclusion and full texts were not accessible for clarification, or abstract or full text not available in English, the articles were excluded (Fig. 1). Two authors, BS and AS, reviewed the articles and decided whether to include or exclude according to the inclusion/exclusion criteria of this article.

2.2. Eligibility criteria

Articles including human participants were accessed and reviewed only. Experimental animal studies, basic science articles, review articles, articles with less than 20 human participants, and meta-analyses were excluded. Articles in which clinical 'hard' cardiovascular outcomes recorded (myocardial infarction, stroke-cerebrovascular events, cardiovascular death, mortality, all-cause mortality, major CVDs, hospitalization due to heart failure, angina, peripheral artery disease (PAD)) and/or clinical tests (EKG evidence for coronary disease, exercise/stress tests, angiographic evidence of coronary artery disease (CAD), blood pressure (BP), flow-mediated dilatation (FMD) of arteries, common carotid artery intima-media thickness (CCA-IMT)) performed were included (Fig. 1).

2.3. Risk of bias

Studies whose objective was not clearly stated were excluded. To minimize risk of confounding factors and bias, articles which had less than 20 participants were also excluded. Moreover, studies not defining the population studied were also excluded. 26 of the 87 studies included were RCTs. In the discussion section of this article, potential biases in the included studies are also discussed.

3. Results

3.1. Study selection and characteristics

PubMed search resulted in 1384 results. After a review as outlined in the inclusion and exclusion criteria, a total of 87 studies were found to be eligible and included in the present review article. Twenty-six of these articles were RCTs, the results of which were also summarized in Table 1.

3.2. Description of reviewed studies

The included articles in each section are presented in chronological order as much as possible. In cases that they used the same dataset or are the continuation of the same study, they were presented consecutively.

3.2.1. Vitamins A-C-E

In a double-blind, crossover study published in 1977, 48 patients with stable angina, positive exercise treadmill test, and angiographic evidence of CAD were given large doses of Vitamin E (1600 IU/day) and placebo for 6 months, separated by 2 months. Drug effectiveness was evaluated by daily angina diaries and maximal exercise treadmill tests. At the end of the study, it was concluded that Vitamin E failed to increase exercise capacity, failed to improve left ventricular function or reduce the frequency of chest pain [5]. This study did not display in the performed PubMed search, but was found through checking references of other studies at that time. The first article on PubMed "Vitamin supplements and cardiovascular health" search dates back to 1982, where the vitamin supplement user, health-conscious 479 elderly Californians were followed up. The standardized mortality ratios of the United States were used to compare any discourse in mortality in this population. Following a 6-year follow-up, it was found that only the cardiovascular standardized mortality ratio was significantly lower in this group of people and 'very high and very low levels' of Vitamin E intake were associated with increased mortality. On the other hand, the authors highlighted that this specific cohort had higher socioeconomic status than the general population and were particularly interested in maintaining a healthy lifestyle [6].

A 1986 prospective study investigating any association between Vitamin C intake and mortality on 3119 people in California over 10 years found that no significant relationship exists between Vitamin C intake and mortality from cancer, CVDs and all-cause mortality. On the other hand, combination of several health habits including never smoking cigarettes, 7–8 h of regular sleep, regular physical activity, moderate-no alcohol use and maintaining proper weight were found to be inversely correlated with mortality [7].

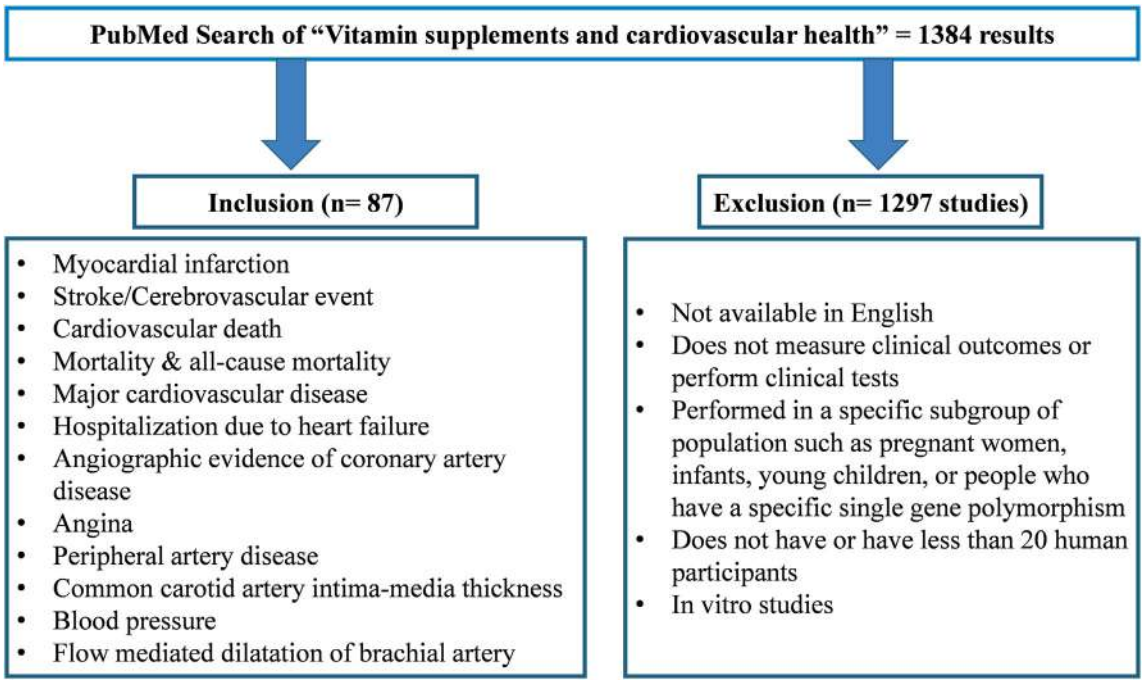

Fig. 1. Chart of inclusion/exclusion criteria.

In 1992, the relationship between Vitamin C intake and mortality was examined in 11,348 US adults aged 25–74 for a median of 10 years (1974–84). In this study, increasing Vitamin C intake was associated with a significantly lower standardized all-cause mortality ratio for males (0.65; 95% CI, 0.52–0.80), females (0.86; 95% CI, 0.55–1.27) and as for CVDs (0.58; 95% CI, 0.41–0.78) for males, and (0.75; 95% CI, 0.55–0.99) for females. In addition, these significant associations persisted after adjusting for age, sex, and other possible confounding factors including cigarette smoking, education, race, and disease history [8]. In 1992, a double-blind, placebo-controlled trial randomized 115 patients enrolled 48 h after percutaneous coronary angioplasty to either 3 × 400 IU/day Vitamin E orally or placebo for 4 months or until symptoms suggesting recurrent stenosis. One-hundred patients completed the protocol and blood tests revealed Vitamin E as the only different parameter, verifying compliance. Restenosis was documented in 35.5% of patients randomized to Vitamin E versus 47.5% in placebo. Overall evidence of restenosis (angiography + stress test) was found to be 34.6% in patients receiving Vitamin E versus 50% in placebo group ($p = 0.06$). Authors self-reported inadequate sample size for the non-significant findings [9].

In a 1993 New England Journal of Medicine *(NEJM)* study where 87,245 female nurses without diagnosed CVDs were asked to fill dietary questionnaires that assessed their consumption of food including Vitamin E. After a follow-up period of 8 years, in this observational study, it was found that people who took Vitamin E supplements had a relative risk (RR) of major CVD of (0.59; 95% CI, 0.38–0.91) and those who were in the upper fifth of the Vitamin E intake cohort had a RR of major coronary disease of (0.66; 95% CI, 0.50–0.87) compared to those who were in the lower fifth of the Vitamin E intake cohort. Since this was not an RCT, it was recommended that the results of the pending RCTs be waited before a public recommendation is made [10].

In 1993, a similar questionnaire-based observational study where 39,910 US male health professionals without diagnosed coronary heart disease (CHD) were enrolled found a lower risk of coronary disease among males with higher intakes of Vitamin E (>60 IU/day vs < 7.5 IU/day) with a multivariate RR of 0.64 (95% CI, 0.49–0.83). In addition, males who took at least 100 IU/day of Vitamin E supplements for at least two years had a lower multivariate RR of coronary disease of 0.63 (95% CI, 0.47–0.84) [11].

In 1996, a questionnaire-based study published in *NEJM*, where 34,486 post-menopausal females were followed-up for approximately 7 years found inverse association between Vitamin E intake and risk of death from CAD. In addition, in this study, a dose–response relationship was also observed. When 21,809 females who did not consume any vitamin supplements were grouped based on Vitamin E intake from the lowest to highest, the RR of CHD was found to range from 1.0 to 0.42 ($p < 0.05$). On the other hand, the intake of Vitamin E from supplements was not found to significantly affect the risk of death from CAD. In addition, no significant association was found between the intake of Vitamin A or C and CAD [12].

A following study found that people who took vitamin supplements were more likely to be physically active, more likely to follow nutritional advices such as avoiding too much salt, sugar; adequate consumption of fiber, vitamins and minerals. Therefore, it was highlighted that supplement use is among a cluster of healthy behavior and studies investigating the effects of dietary supplements ought to control for the confounding health conscious behavior [13].

In 1997, a clinical trial analysis including 285 patients with 'non-insulin dependent diabetes mellitus' (DM), Vitamin C supplementation was not found to be associated with a change in cardiovascular risk factors (systolic − diastolic BP, HDL-C, LDL-C or triglycerides) [14]. In another study; 27,271 Finnish male smokers between the ages of 50–69 years were randomized to either Vitamin E (50 mg), beta-carotene (20 mg), both or placebo for a median follow-up of 6.1 years. This study looked at the potential primary preventive effects of these supplements on major coronary events. The end-points were determined as first major coronary event: either nonfatal MI or fatal CHD. In the end, neither intervention was found to significantly affect these end-points [15].

Table 1
Summary of the RCTs included in this review article.

Author, Year	n	Participants (Age in years)	Supplementation and supplementation details	Results
Stephens NG, 1996 [20]	2002	Patients with angiographically proven lesions **Age:** Mean = 62	α-tocopherol, capsules containing 800 IU daily for the first 546 patients, 400 IU daily for the remaining 489 patients **Follow up duration:** 506 days	Significantly reduced risk of cardiovascular death and non-fatal MI ($p = 0.005$) due to a significant reduction in the risk of non-fatal MI ($p = 0.005$). Non-significant excess of cardiovascular deaths in the α-tocopherol group ($p = 0.61$)
Lee IM, 2005 [25]	39,876	Healthy females **Age:** ≥45 (mean = 54.6)	α-tocopherol 600 IU of α-tocopherol every other day **Follow up duration:** 10 years	Nonsignificant 7% risk reduction ($p = 0.26$) in major cardiovascular events. No significant effects on the incidence of myocardial infarction ($p = 0.96$) or stroke ($p = 0.82$), as well as ischemic or hemorrhagic stroke. Significant 24% reduction ($p = 0.03$) for cardiovascular death. No significant effect on total mortality ($p = 0.53$).
Sesso HD, 2008 [27]	14,461	US male physicians **Age:** >50 (mean = 64.3)	Vitamin E (400 IU synthetic α-tocopherol or its placebo every other day, Vitamin C (500 mg synthetic ascorbic acid or its placebo daily) **Follow up duration:** 8 years	No effect of Vitamin E on the incidence of major cardiovascular events ($p = 0.86$), total MI ($p = 0.22$), total stroke ($p = 0.45$), cardiovascular mortality ($p = 0.43$) and total mortality ($p = 0.15$). No significant effect of vitamin C on major cardiovascular events ($p = 0.91$), total MI ($p = 0.65$), total stroke ($p = 0.21$), cardiovascular mortality ($p = 0.86$) and total mortality ($p = 0.16$). Increased risk of hemorrhagic stroke ($p = 0.04$) associated with Vitamin E.
Toole JF, 2004 [32]	3680	Adults with nondisabling cerebral infarction **Age:** >35	Vitamin B6 (pyridoxine), Vitamin B12 (cobalamine), and folic acid **Follow up duration:** 2 years	10% lower risk of stroke ($p = 0.05$), 26% lower risk of CHD events ($p < 0.001$), and 16% lower risk of death ($p = 0.001$) in the low-dose group. Nonsignificantly lower risk in the high-dose group by 2% for stroke, 7% for CHD events, and 7% for death
Jamison RL, 2007 [37]	2056	Patients who had advanced CKD **Age:** ≥21	Once-daily capsule containing 40 mg of folic acid, 100 mg of pyridoxine hydrochloride, and 2 mg of cyanocobalamin **Follow up duration:** 3.2 years	No significant effect on mortality. No significant effects for secondary outcomes or adverse events: MI, stroke, amputation. No difference in the composite of MI, stroke, and amputations plus mortality ($p = 0.85$), time to dialysis ($p = 0.38$), and time to thrombosis in hemodialysis patients ($p = 0.97$) between the vitamin and placebo groups.
Kwok T, 2012 [46]	50	Vegetarians **Age:** 45 ± 9	Vitamin B12 500 μg/day **Follow up duration:** 24 weeks	Significant improvement of brachial FMD ($p < 0.0001$) and in carotid iMT ($p < 0.05$) No significant change in BP or lipid profile
Peña AS, 2013 [47]	20	Children with type 1 DM, age range of 10–18 **Age:** 15 ± 2	0.5, 2, and 5 mg of folic acid **Follow up duration:** 4 months	No difference in baseline FMD ($p = 0.96$)
van Dijk SC, 2015 [48]	2919	Participants older than 65 years with hyperhomocysteinemia (12–50 μmol/L) **Age:** 74 ± 6.5	Vitamin B12, Folic acid 500 μg Vitamin B12, 400 μg folic acid (Both tablets contained 15 mg (600 international unit) of vitamin D3 to ensure a normal vitamin D status) **Follow up duration:** 2 years	No effect on PWV levels ($p = 0.85$), CCA-IMT (0.76), SBP ($p = 0.35$), DBP (0.07), MI (0.56), any type CVD (0.50), cerebrovascular event (0.17).
Hsia J, 2007 [49]	36,282	Postmenopausal females **Age:** 50-79	200 mg calcium carbonate and 200 IU twice daily Vitamin D3 **Follow up duration:** 7 years	Nonsignificant change in MI or CHD death ($p = 0.50$), CABG or PCI ($p = 0.12$), confirmed angina ($p = 0.30$), hospitalized heart failure ($p = 0.50$), stroke ($p = 0.51$) and TIA($p = 0.13$).
Prentice RL, 2013 [50]	36,282	Postmenopausal females **Age:** 50-79	Calcium carbonate, Vitamin D3, 1000 mg calcium carbonate plus 400 IU of vitamin D3 daily **Follow up duration:** 7 years	No significant difference for MI, CHD, total heart disease, stroke and total cardiovascular disease ($p > 0.05$).
Witham MD, 2012 [53]	58	Community-dwelling patients with baseline Vitamin D deficiency with previous stroke **Age:** Mean = 67	Vitamin D2 (ergocalciferol) 100,000 units of Vitamin D2 (ergocalciferol) **Follow up duration:** 16 weeks	No significant change in office systolic and diastolic blood pressure between groups at 8 weeks (systolic, $p = 0.97$), (diastolic, $p = 0.15$). Significant increase in FMD in the intervention group at 8 weeks ($p = 0.007$) but no significant difference at 16 weeks
Gepner AD, 2012 [56]	114	Post-menopausal women with serum 25(OH)D conc. Between 10 and 60 ng/mL **Age:** Mean = 64	Vitamin D3 Daily oral Vitamin D3 (2500 IU) **Follow up duration:** 4 months	No significant difference between groups in changes in FMD ($p = 0.77$), PWV ($p = 0.65$), or Aortic augmentation index ($p = 0.10$).
Witham MD, 2013 [58]	50	Healthy South Asian women with baseline serum 25(OH)D conc. <75 nmol/L **Age:** Mean = 41	A single dose of 100,000 units oral Vitamin D3 **Follow up duration:** 8 weeks	No improvement in FMD in the vitamin D group relative to placebo at 4 weeks ($p = 0.84$) or 8 weeks ($p = 0.98$). No significant change in any of the other vascular, metabolic or inflammatory outcomes
Witham MD, 2015 [59]	50	Participants with chronic fatigue syndrome **Age:** 49 ± 13	100,000 units of oral Vitamin D3 every two months **Follow up duration:** 6 months	No effect on PWV ($p = 0.93$). No improvement in other vascular and metabolic outcomes, or in the Piper Fatigue scale ($p = 0.73$).
Arora P, 2015 [60]	534	Patients with Vitamin D conc< <25 ng/mL with a SBP of 120–159 mmHg **Age:** 36 ± 10	Vitamin D3 (cholecalciferol) Once-daily oral doses of Vitamin D3 (cholecalciferol), with a total of 4000 IU or 400 IU, in the high-dose and low-dose arms, respectively **Follow up duration:** 6 months	No significant difference in the primary endpoint ($p = 0.71$) or in any of the other secondary endpoints.

Table 1 (*continued*)

Author, Year	n	Participants (Age in years)	Supplementation and supplementation details	Results
Bolland MJ, 2015 [66]	36,282	Post-menopausal women **Age:** 64	Calcium, Vitamin D (not specified) 1 g calcium and 400 IU Vitamin D daily **Follow up duration:** 7 years	No significant change in MI; HR = 1.06, stroke; HR = 0.99, or all-cause mortality; HR = 0.91.
Miskulin DC, 2016 [67]	276	Patients undergoing hemodialysis who had serum levels of 25(OH)D conc. less than 30 ng/mL **Age:** 61 ± 14	Ergocalciferol **Follow up duration:** 6 months	No significant difference in the rates of all-cause and cardiovascular hospitalizations by study arm (p = 0.20 and p = 0.10 respectively). No significant effect on SBP (p = 0.60).
Harris E, 2016 [68]	239	Healthy females aged 50 years or more, and healthy males aged 50–65 years	Multivitamin supplements **Follow up duration:** 4 months	No significant effect for augmentation index (p = 0.743), augmentation pressure (p = 0.718), or central pulse pressure (p = 0.646); nor for peripheral measures of SBP (p = 0.223), DBP (p = 0.223), or pulse pressure (p = 0.223).
Salekzamani S, 2017 [70]	80	Participants who had metabolic syndrome **Age:** Mean = 40	Vitamin D (not specified) 50,000 IU/week **Follow up duration:** 16 weeks	No effect in CCA-IMT (p = 0.732)
Scragg R, 2017 [71]	5110	Participants mostly from family practices in Auckland, New Zealand **Age:** 66 ± 8	Oral Vitamin D3 Initial dose of 200,000 IU followed by 100,000 IU a month **Follow up duration:** 3.3 years	No significant change in CVD and death (p = 0.81). No significant change in secondary outcomes such as MI (p = 0.68), angina (p = 0.13), heart failure (p = 0.34), hypertension (p = 0.45), arrhythmias (p = 0.71), arteriosclerosis (p = 0.54), stroke (p = 0.84) and venous thrombosis (p = 0.45).
Kumar V, 2017 [72]	120	Patients between the ages of 18–70 years with nondiabetic CKD stage 3–4 and Vitamin D conc < 20 ng/mL **Age:** Mean 44	Oral doses of cholecalciferol (300,000 IU) **Follow up duration:** 16 weeks	Significant increase endothelium-dependent brachial artery FMD (P < 0.001). Significant favorable change in PWV (p = 0.001).
Tomson J, 2017 [75]	305	Older people living in the UK **Age:** Mean 72	Vitamin D 4000 IU (100 μg), Vitamin D 2000 IU (50 μg), or placebo daily (Vit D subtype not specified). **Follow up duration:** 12 months	No significant effect on mean levels of BP, HR, or arterial stiffness (p > 0.05)
Sluyter JD, 2017 [76]	517	Adults **Age:** 50–84 (Mean = 65)	Vitamin D3, either Vitamin D3 200,000 IU (initial dose) followed 1 month later by monthly 100,000-IU doses (n = 256) or placebo monthly (n = 261) **Follow up duration:** 1.1. years	Vitamin D group: Non significant improvements in SBP (p = 0.11) and DBP (p = 0.12). Significant improvements in aortic SBP (p = 0.03), augmentation index (p = 0.03), PWV (p = 0.02), peak reservoir pressure (p = 0.01), backward pressure amplitude (p = 0.01).
Manson JE, 2019 [78]	25,871	Males >50 years old and females >55 years old living in the US, of which 5106 were black **Age:** 67 ± 7	Vitamin D3 2000 IU/day and 1 g/day omega-3 fatty acids **Follow up duration:** 5.3 years	No significant difference for major cardiovascular events (p = 0.65).
Wang C, 2009 [81]	128	Obese Chinese females between the ages of 18–55 **Age:** Mean = 42	Multivitamin and mineral supplements, calcium. One tablet of high-dose multivitamin and mineral supplement (MMS), or one tablet of low-dose MMS (Low MMS), or calcium 162 mg **Follow up duration:** 26 weeks	Significant decrease in both SBP and DBP in the MMS group compared to the placebo group (p < 0.05). Non-significant trend of lower DBP at 26-week in the MMS and calcium groups compared to baseline (p < 0.08)
Fulton RL, 2016 [89]	80	Participants who had a history of vascular disease **Age:** 77 ± 5	Vitamin K2 (MK7 subtype) Oral daily 100 μg **Follow up duration:** 6 months	No significant change in endothelial function (p = 0.62). Non-significant improvement in pulse wave velocity (p = 0.15). No significant change in any of the other vascular and physical function outcomes

CABG = coronary artery bypass graft, CCA-IMT = common carotid artery intima-media thickness, CVD = cardiovascular disease, CHD = coronary heart disease, CKD = chronic kidney disease, DBP = diastolic blood pressure, FMD = flow mediated dilatation, MI = myocardial infarction, PCI = percutaneous coronary intervention, PWD = pulse wave velocity, SBP = systolic blood pressure.

Another study enrolling 43,738 males from 40 to 75 years old investigating whether the effects of Vitamin E, Vitamin C, and carotenoid intake can predict the ischemic stroke risk did not find a substantial difference in stroke rates between people whose Vitamin E intake was higher or lower (411 IU/day vs 5.4 IU/day). Moreover, there was also no difference between those who took 700 mg or more Vitamin C vs those who did not use Vitamin C supplements [16].

The Women's Health Study was a randomized, double-blind, placebo-controlled trial where the effects beta-carotene was also tested in the prevention of CVD in 39,876 females over 45 years of age. No difference in CVD was found at a median follow-up of 2.1 years between those who received beta-carotene (19,393 females) or placebo (19,937 females). Therefore, it was concluded that in healthy people, beta-carotene supplementation for a limited period is not associated with any benefit or harm [17].

An observational study utilizing the vitamin intake questionnaire of Cancer Prevention Study II database including 1,063,023 US males and females over 30 years of age found that over a follow-up period of 7 years (1982–1989), males and females taking vitamin supplements overall had a lower mortality due to ischemic heart disease (IHD) and stroke. The inverse association was found to be the strongest in those taking multivitamins and Vitamin A, C, or E compared to those taking A, C, and/or E, and weakest in those taking only multivitamins. On the other hand, no consistent decrease in mortality was found with prolonged (defined as 5–10 years) vitamin intake. Moreover, the people who took vitamin supplements in this cohort were generally more educated, less

overweight, more likely to consume vegetables and drink wine or liquor, which increased the likelihood of confounding factors. After adjusting for potential risk factors besides age, the associations became less evident, and multivariate RRs for IHD were 0.93 (0.87–1.00) for males with IHD taking multivitamins; 0.84 (0.78–0.91) for males without IHD taking multivitamins and Vitamin A, C, or E; and 0.83 (0.77–0.89) in males with IHD taking multivitamins and Vitamin A, C or E. On the other hand, as the authors acknowledged, the study being a very large observational study, confounding factors could not be reliably controlled [18]. A clinical trial examining the effects of Vitamin E supplementation in 520 males and females with hypercholesterolemia (>5 mmol/L, >193 mg/dL) over 6 years found that the average increase in CCA-IMT was 25% slower ($p = 0.034$) in both sexes combined (in the 136 IU Vitamin E plus 250 mg of slow-release Vitamin C, twice daily). On the other hand, the subgroup analyses revealed that the decrease was significant in only males. The effect was found to be higher in participants with either a low baseline Vitamin C levels or existing common carotid artery plaques [19].

In 2002, a randomized controlled trial (RCT) of Vitamin E supplementation in 2002 patients with angiographically proven lesions was published in The Lancet. In this study; 1035 patients were assigned to 800 or 400 IU Vitamin E daily, and 967 were assigned to placebo. After a median follow-up period of 510 days, it was found that Vitamin E supplementation significantly diminished the primary end-point of cardiovascular death and non-fatal MI (RR = 0.53; 95% CI, 0.34–0.83, $p = 0.005$) [20].

Another clinical trial evaluated the association between arterial stiffness and 400 IU Vitamin E supplementation for 10 weeks in postmenopausal females. Only 20 females were enrolled in this randomized, placebo-controlled, crossover trial. It was found that pulse wave velocity (PWV), which was used as a marker of arterial stiffness, did not change. Moreover, at the end of the study, no difference in systolic or diastolic BP was observed between the Vitamin E and the placebo [21].

Using the rationale that Vitamin C can act as a pro-oxidant and glycate proteins under certain in vitro circumstances, a study enrolling 1923 females was designed to look into the relationship between Vitamin C intake and mortality from total CVD, coronary disease, and stroke. The participants who were 'free of CAD' at baseline were followed for 15 years. After adjusting for cardiovascular risk factors, it was found that high Vitamin C intake from supplements (>300 mg/day) was associated with an increased risk of CVD mortality in postmenopausal females with diabetes. However, in participants who did not report diabetes at baseline, no relationship was found between Vitamin C intake and cardiovascular mortality [22].

A randomized, double-blind, placebo-controlled primary prevention trial on the effects of antioxidant vitamins and minerals enrolling 13,017 French adults over a median follow-up of 7.5 years found that a single daily capsule combination of 120 mg ascorbic acid, 30 mg Vitamin E, 6 mg beta carotene, 100 μg selenium, and 20 mg zinc vs placebo did not change ischemic cardiovascular incidence or all-cause mortality [23]. Due to the conflicting results at that time regarding the utility of Vitamin E in the prevention of cardiovascular events, a randomized, double-blind, and placebo-controlled international trial was conducted to evaluate whether long-term supplementation of Vitamin E (400 IU/day) could decrease major cardiovascular events. 3994 people who were at least 55 years old and had vascular diseases and/or DM were enrolled, and followed-up for a median duration of 7 years. The results of this study revealed no difference between major cardiovascular events ($p = 0.34$). Furthermore, patients in Vitamin E supplementation group had a higher risk of heart failure ($p = 0.03$), and a higher risk for hospitalization due to heart failure ($p = 0.045$). Therefore, it was concluded that in patients with DM and/or vascular disease, Vitamin E supplementation does not prevent major cardiovascular events, and could even increase the risk of heart failure [24]. With the same background, an RCT enrolling 39,876 apparently healthy females with a 2 × 2 design was performed. In this study, patients were randomized to 600 IU Vitamin E or placebo every other day. After an average of 10.1 years, no significant difference was found in the incidence of MI, ischemic or hemorrhagic stroke, and total mortality between Vitamin E supplementation and placebo [25].

In the multivariate analysis of a population-based cohort study from central Sweden, which had healthy 38,994 males participants between the ages of 45–79 over an average of 7.7 years, found no association between the use of multivitamin, Vitamin C or Vitamin E supplementation and all-cause mortality or CVD in participants who reported adequate diet. On the other hand, among males who reported insufficient diet at baseline, statistically significant inverse association between dietary supplements and CVD was found (RR = 0.72; 95% CI, 0.57–0.91) [26].

Another 2008 JAMA RCT looked into whether long-term supplementation of Vitamin C or E could decrease the risk of cardiovascular events among males. In this study; 14,641 US male physicians, who were over 50 years of age were enrolled. After a mean follow-up period of 8 years and supplementation of 400 IU of Vitamin E every other day and 500 mg of daily Vitamin C; neither Vitamin C nor Vitamin E supplementation was found to be associated with a significant difference in the incidence of major cardiovascular events, total MI, total stroke, and cardiovascular mortality. On the other hand, Vitamin E supplementation was found to be associated with a significantly increased risk of hemorrhagic stroke ($p = 0.04$, HR = 1.74; 95% CI, 1.04–2.91) [27].

A study aimed to investigate the association between individual-level dietary intakes of antioxidant Vitamin C, E, beta-carotene; and all-cause mortality. The investigators used data from Health, Alcohol and Psychosocial factors in Eastern Europe cohort study. In this cohort, a baseline survey was in 2002–2005, and 28,945 males and females between the ages of 45–69 were enrolled. Even though some specific group comparisons yielded statistical significance, it was concluded that in this population with low-vitamin supplementation; no strong, dose–response evidence was found for the protective effects of vitamin intake [28].

3.2.2. Vitamins B6–B9–B12

A large study utilizing the food questionnaire from Nurses' Health Study (80,092 females) investigated the effects of folate and Vitamin B6 on nonfatal MI and fatal CHD. Over a follow-up period of 14 years, after controlling for several potential confounding factors such as smoking, alcohol intake, and hypertension, comparison of extreme quintiles had a RR confidence interval of 0.55–0.87 for folate (median intake, 696 μg/d vs 158 μg/d) and 0.53–0.85 for Vitamin B6 (median intake, 4.6 mg/day vs 1.1 mg/day); higher intake was associated with a lower RR of in both cases [29].

A prospective study with a follow-up period of 12 years published in 2003 examining the relationship between PAD and folate, Vitamin B6, and Vitamin B12 intake in 51,529 male healthy professionals in the US found that for every 400 μg/day increase in folate intake, the multivariate adjusted PAD risk decreased by 21%. Furthermore, males who were in the top group of folate intake (median 840 μg/day), had a 33% lower risk of PAD compared to males in the bottom group (median 244 μg/day). On the other hand, even though there seemed to be an inverse relationship between Vitamin B6 intake and PAD, this change was not conclusive ($p = 0.06$), and non-existent for Vitamin B12 ($p = 0.12$) [30].

A study enrolling 43,732 males aged 40 to 75, free of CVD and diabetes at baseline was published in 2004. This study examined

the association between Vitamin B6, Vitamin B12, folate intake, and ischemic or hemorrhagic stroke. At the end of a 14-year follow-up period, it was found that high folate intake was associated with a significantly lower multivariate RR of ischemic stroke = 0.71 ($p = 0.05$), but similar rates of hemorrhagic stroke. On the other hand, this study was based on food questionnaires and participants with higher dietary folate intake were also found to take more vitamin supplements [31]. Therefore, it was not possible to draw conclusions regarding the effects of vitamin supplements per se on stroke risk. A 2004 double-blind RCT enrolling 3680 adults with nondisabling cerebral infarction randomized patients to high or low formulations of Vitamin B6, Vitamin B12, and folate with a follow-up period of 2 years. At the end of the study, whereas it was found that homocysteine levels decreased more in the high-dose vitamin supplementation group, this difference did not translate into a clinical end-point (similar rates of recurrent cerebral infarction, CHD, and death) [32].

In a 2005 which enrolled patients with heart failure and elevated plasma homocysteine levels (>15 μM) with a median age of 81 years tested whether the daily supplementation of 3 mg Vitamin B6, 0.8 mg folate, and 0.5 mg Vitamin B12 for 6 weeks could have any significant change in associated parameters. Similar to other studies, the vitamin supplementation was able to significantly reduce plasma homocysteine levels from 17.9 ± 0.6 μM to 13.8 ± 0.1 μM ($p < 0.01$). Moreover, consistent with other studies; in this study, the authors found that even though plasma homocysteine levels decreased in the supplementation group, inflammatory markers remained constant. On the other hand, in contrast to other studies; in this study, vitamin supplementation was shown to be associated with a decreased mean arterial BP. It is likely that the reason why supplementation was found to have a clinically translatable change in this study could be due to the enrollment criteria and the baseline characteristics of patients (e.g. baseline subnormal levels of supplemented vitamins) or simply related to confounding factors (no placebo group), considering that the decrease in mean arterial BP was only 5 mmHg, from 95 mmHg to 90 systolic, and from 75 mmHg to 70 diastolic [33].

In 2006, a placebo-controlled study with hard primary outcomes was published in *NEJM*. In this study; 5522 patients who were older than 55 years of age and had a vascular disease or diabetes were randomized to either the combination of 2.5 mg folic acid, 50 mg Vitamin B6, and 1 mg Vitamin B12 or placebo. The average follow-up duration was 5 years. Compared to placebo, vitamin supplementation did not result in a significant decrease in the risk of death due to cardiovascular events (RR = 0.96; 95% CI, 0.81–1.13), MI (RR = 0.98; 95% CI, 0.85–1.14). On the other hand, while patients in the supplementation group had a lower RR of stroke (RR = 0.75; 95% CI, 0.59–0.97), vitamin supplementation was also found to be associated with an increased hospitalization due to unstable angina (RR = 1.24; 95% CI, 1.04–1.49) [34,35].

The Atherosclerosis and Folic Acid Supplementation Trial to determine whether high-dose daily folic acid (15 mg) could slow to the progression of atherosclerosis and reduce cardiovascular events in patients with chronic renal failure enrolled 315 patients with a mean age of 57. After a median follow-up of 3.6 years, plasma homocysteine levels were found to be decreased by 19% in the folic acid group. Despite this, no difference was found in the rate of carotid IMT change. In addition, there was also no significant difference in MI, stroke, and cardiovascular death between the folic acid and the placebo groups [36].

A double-blind RCT enrolling 2056 patients who had advanced chronic kidney disease (CKD) to determine whether high doses of folic acid and Vitamin B6–B12 could decrease the mortality in patients was conducted between 2001 and 2006. Participants were given a daily capsule containing 40 mg folic acid, 100 mg Vitamin B6, 2 mg Vitamin B12 or placebo. Baseline homocysteine levels were comparable in each group. After a median follow-up of 3.2 years, in the intervention group, homocysteine levels were found to be significantly lower. On the other hand, supplementation was not associated with a change in mortality (HR = 1.04; 95% CI, 0.91–1.18). Moreover, no difference was found between the supplementation group and placebo with respect to MI or stroke rates [37].

A cohort of apparently healthy 40,803 participants from Japan between 40 and 59 years were studied through a food questionnaire. In this study, data regarding vitamin supplementation were not specified. On the other hand, people who had a lower dietary intake of Vitamin B6 had a significantly higher risk of CHD and MI [38].

A 2008 trial looking into composite outcomes of MI, stroke, coronary revascularization or cardiovascular mortality published in *JAMA* enrolled 5442 females with either a history of CVD or 3 or more coronary risk factors and randomized them into either receiving a daily combination pill of 2.5 mg folic acid, 50 mg Vitamin B6, and 1 mg Vitamin B12 or a placebo. Participants were followed up for 7.3 years. At the end of the follow-up period, it was found that participants who received vitamin pills had their geometric mean plasma homocysteine levels decreased by 18.5% ($p < 0.001$). On the other hand, this difference did not translate into clinical end-points and no difference was found between placebo and vitamin supplementation group with regard to composite CVD primary end-point ($p = 0.65$, RR = 1.03; 95% CI 0.90–1.19). There was also no difference in secondary outcomes including MI, stroke, and CVD mortality [39].

Homocysteine levels have been shown to be a risk indicator for CVDs. On the other hand, it was not shown whether decreasing homocysteine levels through supplementation could have an effect on hard end-points, such as a decrease in the incidence of CVD. A clinical trial in France with a factorial design examining this hypothesis was designed in 2003, and published in 2010. In this study, investigators enrolled 2501 people who had a history of MI, unstable angina, or ischemic stroke in the past year. The patients were given 5-methyl tetrahydrofolate (560 μg), Vitamin B6 (3 mg), Vitamin B12 (20 μg) and/or n-3 fatty acids with a follow-up period of 5 years [40,41]. The results indicated that even though Vitamin B supplementation decreased plasma homocysteine levels by 19%, it had no significant effect on major vascular events. Moreover, similarly, omega-3 fatty acid supplementation increased plasma concentration of omega-3 by 37%, but had no significant effect on major vascular events. Therefore, in this trial with a 5-year follow-up, it was concluded that Vitamin B supplementation or omega 3 fatty acid supplementation does not have clinically translatable effects on CVD when introduced in the following year after major cardiovascular events [42]. Using the data obtained from the same study, it was concluded that these supplementations also do not result in a significant change in BP [43].

A randomized trial, this time to test whether long-term supplementation of 2 mg folic acid and 1 mg daily Vitamin B12 could decrease the risk of CVD enrolled 12,064 survivors of MI in hospitals. After 6.7 years of follow-up, homocysteine levels were found to be significantly lower in the supplementation group compared to placebo. On the other hand, the supplementation was not associated with significantly different rates of major coronary events or strokes. Furthermore, no difference was found between the two groups in deaths attributable to vascular or non-vascular causes [44].

A sub-study of Western Norway B Vitamin Intervention Trial (WENBIT) randomized 348 patients who had percutaneous coronary intervention to daily oral treatment with folic acid, Vitamin B12, and Vitamin B6 or placebo in a 2 × 2 factorial design. After a

median follow-up of 10.5 months, total plasma homocysteine levels were found to decrease by 22% in the supplementation group. On the other hand, the comparison of baseline and post-intervention coronary angiograms was not found to be associated with a change in stenosis diameter or lumen diameter [45].

A crossover double-blind RCT of Vitamin B12 supplementation recruited 50 vegetarians (for at least 6 years). These participants were given Vitamin B12 (500 μg/day) for 12 weeks or placebo, with a wash-out period of 10 weeks before crossover, followed by 24 more weeks of supplementation. 70% of the participants had a serum Vitamin B12 level which was below 150 pmol/L. Following 24-weeks of open label Vitamin B12 supplementation, significant improvements in both FMD of brachial artery and carotid IMT were found, which were not observed in the placebo group. Therefore, it was concluded that people who have subnormal Vitamin B12 levels, Vitamin B12 supplementation may be helpful in the improvement of arterial function and the prevention of atherosclerosis [46].

A double-blind, crossover RCT evaluating the lowest effective dose–response of folic acid on endothelial function in children with type 1 DM was conducted in Australia. 20 children with an age range of 10–18 were enrolled and each child received 0.5, 2, and 5 mg of folic acid and placebo, in random order. The control group were selected among children with type 1 diabetes before folate fortification in Australia. As a primary outcome, authors looked into FMD as a measure of endothelial function. In the interventional trial, no difference was observed among the 20 participants in FMD. On the other hand, comparison of these participants and the control group from pre-folate fortification era found significant improvements in glyceryl trinitrate-mediated dilatation [47].

In 2015, 2-year results of B-PROOF trial was published. This study was a double-blind RCT which enrolled 2919 participants older than 65 years with hyperhomocysteinemia (12–50 μmol/L) and randomized these participants to 500 μg Vitamin B12, 400 μg folic acid, or placebo for 2 years. Even though in the supplementation group serum homocysteine levels were significantly lower, analysis of the effects of supplementation on arterial stiffness assessed by pulse-wave doppler did not yield any significant results. Moreover, carotid IMT comparisons did not result in any difference [48].

3.2.3. Vitamin D

An RCT investigating the association between Vitamin D + calcium supplementation on cardiovascular events randomized 36,282 postmenopausal females from 50 to 79 years of age to 500 mg calcium carbonate and 200 IU twice daily Vitamin D found that, after 7 years of follow-up, calcium and Vitamin D supplementation was not associated with an increase or a decrease in cardiovascular events [49,50]. On the other hand, a subgroup analysis of the same cohort suggested that Vitamin D supplementation could be beneficial among postmenopausal females without major heart failure precursors [51].

A clinical trial enrolling 28,886 participants from the US over 45 years of age based on a semiquantitative food questionnaire revealed that the risk of hypertension decreased in participants with a higher dietary calcium intake (multivariate RR = 0.87) and Vitamin D intake (multivariate RR = 0.95). On the other hand, the risk of hypertension did not change with calcium or Vitamin D supplementation [52].

An RCT to test whether Vitamin D supplementation could reduce BP and improve markers of vascular health in patients who previously had a stroke, enrolled community-dwelling patients with a baseline 25-hydroxyvitamin D levels of <75 nmol/L (<30 ng/mL). 58 patients (mean age 67) were randomized to either 100,000 units of Vitamin D2 or placebo at baseline. At the end of 8 weeks, office systolic and diastolic BPs were similar between the two groups. Furthermore, FMD of brachial artery was not significantly different at 16 weeks [53].

A prospective evaluation of dietary + supplementary Vitamin D use and CVD incidence was investigated from the Osteoporotic Fractures in Men (MrOS) study. Vitamin D intakes of 813 males with a mean age of 76.4 were evaluated via a food frequency survey. The median 25(OH) Vitamin D level was 25.3 ng/mL. After a median follow-up of 4.4 years, after adjusting for potential confounding factors, no difference was found between those in the highest intake quartile vs lowest. Furthermore, no difference in CVD incidence was found between Vitamin D deficiency (<15 ng/mL) and Vitamin D sufficiency (>30 ng/mL) (HR = 1.34; 95% CI, 0.65–2.77) [54]. Another analysis of this cohort in 2014 reached a similar conclusion [55].

An RCT of whether Vitamin D supplementation could decrease cardiovascular risk randomized 114 participants to either daily Vitamin D3 (2500 IU) for 4 months, or placebo. At the end of the trial, no difference was found in the comparison of baseline and after intervention results between brachial-artery FMD, carotid-femoral PWV, and aortic augmentation index [56]. Therefore, this study too, joined the dozens of studies with a negative result in this subject.

A study enrolled 90 participants who had Vitamin D deficiency (<20 ng/mL) and randomized them to 50,000 IU ergocalciferol (weekly) or placebo for 12 weeks. As well as many other parameters; a clinically significant parameter, BP, was not found to significantly differ at the end of the study even though the participants had Vitamin D deficiency at baseline [57].

A randomized parallel group, double-blind, placebo-controlled trial enrolled 50 healthy South Asian females who had baseline 25-hydroxyvitamin D levels <75 nmol/L (<30 ng/mL). These females were randomized to receive a single dose of 100,000 units of oral Vitamin D3 or placebo. Primary outcome was determined as endothelial function determined by brachial artery FMD. Some of the secondary outcomes included BP, arterial stiffness, and microvascular function. Even though mean baseline 25-hydroxyvitamin D level was as low as 27 nmol/L (10.8 ng/mL), no improvement was found in FMD or BP at 4 weeks or at 8 weeks [58].

A double-blind, parallel-group RCT randomized as many as 50 participants with chronic fatigue syndrome to receive 100,000 units of oral Vitamin D3 or placebo every 2 months, for 6 months. At the end of the trial, no effect was observed on the primary outcome of carotid-femoral PWV. In addition, the secondary outcome of FMD of the brachial artery did not show improvement [59]. A 2015 double-blind RCT tested whether supplementation of Vitamin D could be useful to reduce BP. To test this hypothesis, 534 patients with low Vitamin D level (<25 ng/mL) with a systolic BP of 120–159 mmHg were enrolled. Participants were randomized to oral high-dose (4000 IU/day) or low-dose (400 IU/day) for 6 months. Median 25-hydroxyvitamin D level at baseline was 15.3 ng/mL. At the end of the study period, no significant difference was found in mean 24-h systolic BP, ambulatory diastolic BP, clinic systolic and diastolic BPs [60].

In 2015, a study to evaluate whether Vitamin D supplementation could affect 24-h systolic ambulatory BP enrolled 200 participants who had 25-hydroxyvitamin D levels <30 ng/mL and arterial hypertension in Austria. The participants were randomized to receive daily Vitamin D3 (2800 IU) oil drops, or placebo (100 participants each). At the end of 8 weeks, Vitamin D supplementation had no significant effect on BP [61].

A Japanese study to investigate multivitamin use and the risk of stroke enrolled 72,180 Japanese males and females who were free from cardiovascular disease and cancer. After a median follow-up of 19.1 years, multivitamin use was found to be associated with a

trend for lower mortality due to stroke (HR = 0.87; 95% CI, 0.76–1.01). Sub-analysis of strokes found that this trend was attributable to a trend for a decrease in ischemic stroke. Furthermore, a significant association was found between multivitamin use and lower risk of mortality from stroke in people who had less than 3 fruits/vegetables in a day (HR = 0.80; 95% CI, 0.65–0.98). Therefore, it was concluded that multivitamin use was associated with a lower risk of mortality due to stroke, especially in those who consumed a diet poor of fruits and vegetables [62].

A following study prospectively examined 19,635 males from the Physicians' Healthy Study. Mean age was 66.4 years and only 2.3% of participants used Vitamin D supplements. After an average follow-up of 9.3 years, no association was found between dietary Vitamin D intake and heart failure [63].

A double-blind, randomized trial to test whether Vitamin D supplementation could improve BP in Native American females enrolled 98 participants with a mean age of 61 years. The participants were randomized to daily Vitamin D 400 IU or 2500 IU for 6 months. At the end of the study period, no difference was found in the comparison of baseline and post-intervention BP. Furthermore, no significant difference in BP, central pulse pressure, or aortic augmentation index was observed between 400 IU or 2500 IU intervention arms [64].

A study looking into possible associations between calcium-Vitamin D supplementation, serum concentrations of calcium, 25-hydroxyvitamin D; and subclinical CVD phenotypes such as IMT, ankle-brachial index (ABI), intermittent claudication, and atrial fibrillation (AFib) examined the data from 1601 participants between the ages of 50–81 in Germany. Even though higher 25-hydroxyvitamin D serum levels were associated with a lower incidence of asymptomatic PAD detected by ABI, no significant association was found between supplementation or CVD phenotype [65].

Another study in 2015 used the opportunity created by Women's Health Initiative Calcium and Vitamin D RCT. In this trial, participants were randomized to 1 g calcium and 400 IU Vitamin D daily or placebo for 7 years. On the other hand, participants were also given permission to use personal calcium and Vitamin D supplements. Therefore, in addition to the comparison of randomized groups, the data for an observational analysis was also possible in this trial. Comparison of supplementation (n = 18,176) vs placebo (n = 18,106) within the entire cohort did not find any significant association between supplementation and the incidence of MI, stroke or death. Moreover, once participants who were taking supplements for personal reasons were excluded, this non-significant association persisted (n = 7891 for Vitamin D/calcium supplementation and 7755 for placebo) [66].

In a 2016 RCT, 276 patients undergoing hemodialysis who had serum levels of 25-hydroxyvitamin D less than 30 ng/mL were randomized to ergocalciferol or placebo for 6 months. This trial was not adequately powered to test the secondary outcome of cardiovascular hospitalizations and had no-effect on clinical end-points [67].

Another RCT, also published in 2016 enrolled 160 females and 79 males between the ages of 50–65 years old who were free of CVD. In this RCT, participants were randomized to multivitamin supplements or placebo. After 4 months, no treatment effect was observed in systolic, diastolic or pulse pressures in these healthy participants [68].

A cross-sectional study including 1054 adults aged more than 40 years old from rural South Korea was performed to investigate the association between dietary intake of Vitamin D and metabolic/arterial changes including BP, brachial ankle PWV, and carotid IMT. As opposed to many other studies, interestingly, in this study, dietary Vitamin D was inversely associated with diastolic BP and brachial ankle PWV among males. In addition, once multinutrient supplement users were excluded, this association disappeared [69].

An RCT conducted in Iran enrolled 80 participants who had metabolic syndrome and randomized the participants into either Vitamin D (50,000 IU/week) or placebo for 16 weeks. Even though laboratory values such as IL-6, vascular endothelial adhesion molecule-1, and E-selectin had significantly improved in Vitamin D supplementation group compared to baseline, no difference was observed in CCA-IMT at the end of the 16 week supplementation [70].

In 2017, the result of Vitamin D Assessment Study was published in *JAMA Cardiology*. This study was conducted to test whether the previously published negative results of RCTs of Vitamin D supplementation was due to the use of too low Vitamin D dose. Therefore, in this study, investigators randomized 5110 participants mostly from family practices in Auckland, New Zealand to oral vitamin D3 (n = 2558) with an initial dose of 200,000 IU followed by 100,000 IU a month or placebo (n = 2552) for 3.3 years. The mean age of the participants was 65.9 years. Baseline 25(OH)D level was 26.5 ng/mL. At the end of the study, similar rates of CVD were observed between Vitamin D supplementation and placebo group (HR = 1.02; 95% CI, 0.87–1.20). Importantly, similar rates of cardiovascular events were also observed in participants who were found to be Vitamin D deficient at baseline [71].

A positive result was reported in a 2017 RCT, where 120 patients between the ages of 18–70 years with nondiabetic CKD stage 3–4 and Vitamin D deficiency (<20 ng/mL) were randomized to two directly observed oral doses of cholecalciferol (300,000 IU) or placebo at baseline and at 8 weeks. At the end of 16 weeks, cholecalciferol supplementation was found to be associated with a significant improvement in endothelium-dependent brachial artery FMD [72].

A randomized, double-blind trial looked into the effects of calcitriol vs cholecalciferol supplementation on vascular endothelial function in patients with CKD and Vitamin D insufficiency (<30 ng/mL). 128 patients were randomized to oral cholecalciferol (2000 IU/day) or calcitriol (0.5 μg/day) for 6 months. 115 participants completed the study and after 6 months, neither calcitriol nor cholecalciferol was found to improve brachial artery FMD [73].

A study from Iran enrolled 998 apparently healthy adolescent girls in Iran. All participants were given 50,000 IU/week for 9 weeks. Vitamin D deficiency was found to be 90% at baseline, which was reduced to 16.3% after 9 weeks of Vitamin D supplementation. Furthermore, at the end of study, a significant reduction in diastolic BP was found. On the other hand, systolic BP was comparable to baseline levels [74].

Biochemical Efficacy and Safety Trial of Vitamin D was a double-blind RCT of 305 older people living in the UK. The participants were randomized to daily Vitamin D (4000 IU) (2000 IU) or placebo. At 12 months, it was found that randomization to Vitamin D supplementation groups had no significant effect on mean BP, heart rate or arterial stiffness. Moreover, no difference was found in echocardiographic measures of cardiac function or NT-proBNP concentration [75].

A study to evaluate the effects of monthly high-dose Vitamin D supplementation on BP was undertaken. In this double-blind RCT 517 adults were recruited. 256 participants were randomized to an initial 200,000 IU of Vitamin D3, followed 1-month later by 100,000 IU and 261 patients were randomized to placebo. In this trial, subgroup analyses found that those who had baseline Vitamin D levels of less than 25 ng/mL, the supplementation had a trend towards decreasing BP. Brachial systolic BP, brachial diastolic BP had a lower, but non-significant mean values after supplementation. On the other hand, statistical significance was reached for aortic systolic BP, augmentation index, peak reservoir capacity, backward

pressure amplitude, and for PWV. Despite these positive results, in the full cohort, these effects were not evident. That is, the improvements were restricted to the group which had lower 25-hydroxyvitamin D levels at baseline [76].

In 2018, data from the UK Biobank cohort comprising 502,637 males between 40 and 69 years of age were used to investigate the association between calcium/Vitamin D supplementation and cardiovascular events/deaths in UK. The supplementation data were self-reported by the participants. A total of 475,255 participants with a median age of 58 years had complete data on supplementation, of which 19,089 reported consumption of Vitamin D supplements. After adjustments, no significant association was found between Vitamin D intake and admission for MI and IHD. The authors concluded that there was no evidence that the use of calcium/Vitamin D supplementation was associated with an increased risk of hospital admission or death after cardiovascular events [77].

The results of a major RCT of 2 × 2 factorial design of Vitamin D3 supplementation was published in *NEJM* in 2019. The investigators enrolled 25,871 participants, of which 5106 were black, and randomized them to Vitamin D3 2000 IU/day and 1 g/day omega-3 fatty acids for the prevention of cancer and CVD. The participant were males >50 years old and females >55 years old living in the US. After a median follow-up of 5.3 years, major cardiovascular events were not different between Vitamin D supplementation and placebo group, p = 0.69 (HR = 0.97; 95% CI, 0.85–1.12). Moreover, expanded composite end-point of major cardiovascular events plus coronary revascularization was also comparable between the groups (HR = 0.96; 95% CI, 0.86–1.08). Furthermore, stroke rates were also similar between the groups (HR = 0.95; 95% CI, 0.76–1.20) as well as death from cardiovascular causes (HR = 0.99; 95% CI, 0.87–1.12). Therefore, this very impactful trial concluded that supplementation with Vitamin D is not associated with a lower incidence of cardiovascular events [78]. A study investigating the efficacy of calcium/Vitamin D supplementation on the prevention of AFib was undertaken by the secondary analysis of Women's Health Initiative randomized trial. Among 16,801 participants, over an average of 4.5 years, no significant difference in incident AFib was found between placebo and calcium/Vitamin D supplementation arms (HR = 1.02; 95% CI, 0.92–1.13) [79].

In 2020, a randomized, placebo-controlled trial of Vitamin D supplementation and cardiovascular health was published. Importantly, this study did not enroll healthy adults, but the participants here were obese children between 10 − 18 years old. In this trial, investigators randomized participants to 1000–2000 IU/day or 600 IU/day Vitamin D3. At 3-month and 6-month follow-up, no difference in endothelial function, arterial stiffness, and systolic BP was found between 1000–2000 IU supplementation and 600 IU supplementation. On the other hand, systolic and diastolic BP were significantly lower in the 1000 IU/day group compared to 600 IU/day group [80].

3.2.4. Multivitamins and vitamin K

An RCT published in 2009 investigated the effects of multivitamin and multimineral supplementation in 128 obese Chinese females between the ages of 18–55. In this double-blind RCT, after 26 weeks of supplementation, systolic and diastolic BP were found to be significantly lower in the high-dose multivitamin and mineral supplementation group compared to the placebo arm. Interestingly, in the same study, another group which had about half the amount of vitamin and mineral supplementation did not have a significant difference in systolic or diastolic BP at the end of 26 weeks compared to the placebo arm, which is most likely due to the small differences in baseline between the groups as shown in Fig. 2 of that study. Moreover, the comparison of baseline and at end-point comparison of systolic and diastolic BPs were similar within the groups. Furthermore, the study did not look into any clinical end-points over a long period of time to justify a recommendation in this subgroup of obese Chinese females [81].

A 2009 multicenter study utilizing the data from Women's Health Initiative clinical trials and an observational study enrolled 161,808 participants. 41.5% of participants reported use of multivitamins. After a follow-up period of about 8 years, multivariate-adjusted analyses did not reveal any significant association between multivitamin intake and CVD, MI, stroke or mortality [82].

Multivariate adjusted analysis of Japan Public Health Center-based prospective cohort comprising 28,903 males and 33,726 females, whose consumption of vitamin supplements was gathered via surveys between 1990-1994 and 1995–1998 found no significant association between the use of vitamin supplementation and the risk of CVD in males. On the other hand, consistent use was associated with a significantly lower CVD risk (HR = 0.60; 95% CI, 0.41–0.89). Despite this improvement, past and recent use of vitamin supplements was also associated with significantly higher risk of cancer. Considering this was an observational cohort without randomization or placebo control, the authors suggested that preexisting underlying conditions may have confounded the results [83].

In 2012, the authors investigated the data from EPIC-Heidelberg cohort of 23,943 participants without pre-existing cancer or MI/stroke. After an average follow-up of 11 years, the authors found that baseline users of antioxidant vitamin supplements had a significantly lower all-cause mortality (HR:0.58, 95% CI, 0.38–0.88). On the other hand, after adjustment for potential confounding factors, no association was found between vitamin/mineral supplements, multivitamin supplements, and all-cause mortality. Furthermore, the comparison of those who never used vitamins and those who started using vitamin/mineral supplements during the study period revealed increased all-cause mortality for those who started supplementation (HR:1.58; 95% CI, 1.17–2.24) [84].

A multicenter, double-blind, placebo-controlled, 2 × 2 factorial trial enrolled 1708 patients aged 50 or older who had MI at least 6 weeks earlier and had creatinine levels of 176.8 mol/L (2.0 mg/dL) or less. The participants were randomized to either 28-component high-dose multivitamin and multimineral mixture or placebo. After a median follow-up of 55 months and vitamin supplementation for a median of 31 months, no difference in total death, recurrent MI, stroke, coronary revascularization or hospitalization for angina was found between the supplementation and placebo group [85].

Another prospective study to investigate a possible association with multivitamin use and short/long term CVDs used the data provided by 37,193 females in the Women's Health Study. These participants were over 45 years of age, free of cancer and CVD at baseline. After an average of 16.2 years, multivitamin use was not associated with higher or lower major cardiovascular events, stroke or cardiovascular death [86].

A study investigating the multivitamin-mineral and multivitamin use and CVD specific mortality among US adults without CVD used restricted data from NHANES III study (1988–1994, n = 8678, age 40 and above). The investigators used the national death index and examined for any difference between national death index and data extracted from NHANES III. The authors reported no significant association between CVD mortality and multivitamin-mineral or multivitamin use compared with non-users. On the other hand, when looked at those who reported the length of supplement use >3 years, a significant association was found only for females, but not for males. Despite this, no significant association was found between supplement use and cardiovascular mortality in fully adjusted models [87].

A large case–control study enrolled 2506 patients with venous thrombosis, 2506 partner controls, and 2684 random-digit dialing

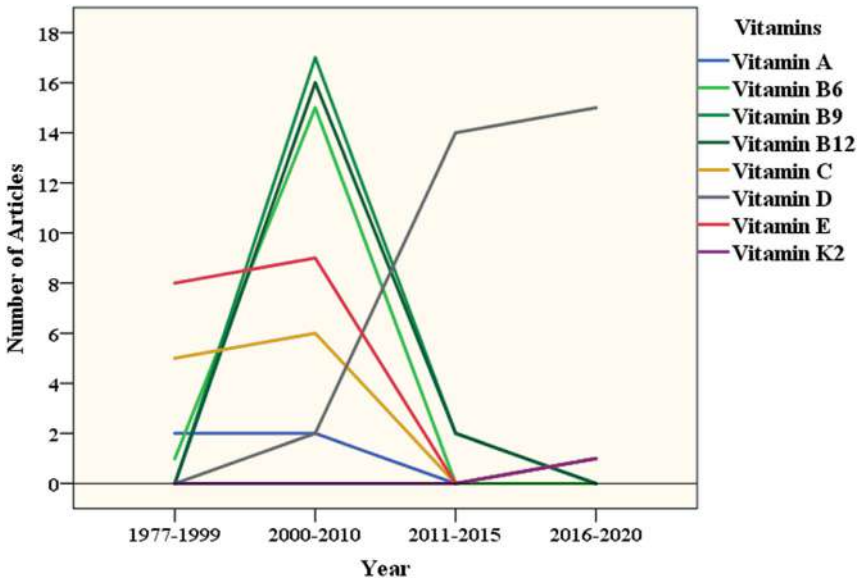

Fig. 2. Line graph showing the publication trend for each vitamin supplementation article by years.

controls to investigate whether vitamin supplementation could decrease the risk of venous thrombosis. It was reported that those who consumed vitamin supplementation had 37% lower risk of venous thrombosis compared to non-vitamin users. On the other hand, when patients were compared with partner-controls, this difference was nullified. Therefore, it was reported that after extensive adjustments, vitamin supplementation was not associated with a significantly decreased risk of venous thrombosis, and the initially found statistical significance was attributed to confounding factors [88].

A double-blind RCT randomized 80 participants who had a history of vascular disease to 6 months of oral daily 100 μg Vitamin K2 or placebo. Mean age of the participants was 77. At the end of the study, the change in carotid-radial PWV was not significant and no significant change was detected in endothelial function [89].

Another study was conducted to investigate whether multivitamin use is associated with the risk of CVD in healthy males at baseline studied the data from the Physicians' Healthy Study I who were free of CVD or cancer at baseline. After a median follow-up of 12.2 years, multivariate-adjusted analyses did not reveal any significant difference in major cardiovascular events between baseline multivitamin users and non-users (HR = 0.84; 95% CI, 0.84–1.05). Baseline vitamin use was also found to be inversely associated with the risk of cardiac revascularization (HR = 0.86; 95% CI, 0.75–0.98). Furthermore, a self-reported duration of more than 20 years of multivitamins was found to be significantly associated with a lower risk of major CV events (HR = 0.56; 95% CI, 0.35–0.90) [90]. Because little was known at that time regarding the effects of multivitamins on the development of hypertension, the investigators studied the data of 28,157 females from the Women's Health Study who were at least 45 years of age and were free from CVD, hypertension, or cancer at baseline. Development of hypertension was tracked during the follow-up period. After a median follow-up of 11.5 years, the investigators found no association between neither baseline nor time-varying multivitamin use and the risk of hypertension development. Duration of multivitamin use was also not associated with the development of hypertension. Therefore, it was concluded that multivitamin use was not associated with an increased or decreased risk of development of hypertension [91].

Another study with a 32 year follow-up looked into a possible association between multivitamin use and stroke incidence and stroke mortality. 86,142 females from Nurses' Health Study between the ages of 34–59 years who were free of diagnosed CVD at baseline were included in this study. At the end of the follow-up period (from 1980 to 2012), multivariate analyses found that females who were currently using multivitamins did not have a lower risk of total stroke compared to non-users (RR = 1.02; 95% CI, 0.93–1.11). Moreover, no difference in stroke incidence was observed in comparison between those who used multivitamins for more than 15 years or those who reported a lower quality diet (defined as Alternative Healthy Eating Index below the 25th percentile) [92].

In 2019, another analysis of the National Health and Nutrition Examination Survey was published. 30,899 adults over 20 years of age participated in this survey. In this analysis, investigators looked at the association between supplement use, nutrient intake from foods and supplements, and mortality among US adults. After a median follow-up of 6.1 years, while ever-use of dietary supplements was not found to be associated with mortality; adequate intake of Vitamin A, Vitamin K, magnesium, zinc, and copper was associated with a lower all-cause CVD mortality. Importantly, these associations were restricted to nutrient intake from foods. Therefore, the authors concluded that supplement use was not associated with a mortality benefit [93].

4. Discussion

In the present review article, all studies (87) carried out to date which fit the inclusion/exclusion criteria of this review were reviewed (Fig. 1). Early studies, which were not randomized clinical trials, found strong correlations between improved health and vitamin supplementation and/or serum vitamin levels. These correlations were not only limited to improved markers such as decreased blood pressure control or improved lipid profiles, but some studies also implied that vitamin supplementation or higher serum vitamin levels might improve clinical end-points. For example, in 1987, a survey based observational study investigating Vitamin A, C, E levels and mortality rates in several European countries revealed that regions with medium-to-high coronary mortality had a significantly lower

Vitamin C level. In addition, in areas with low-to-medium coronary mortality, plasma Vitamin E levels were found to be significantly higher (11.5 mg/L vs 9 mg/L) [94]. Another study in 1987 which followed 10,532 people from the Netherlands over a 9-year period found no association between the serum levels of Vitamin A or E in patients died of CVDs and their age and gender matched controls [95]. In 1997, a clinical trial investigating the effects of folic acid supplementation on plasma homocysteine levels of 30 healthy male volunteers found that 200 μg or 400 μg of daily folic acid supplementation for 6–14 weeks was able to decrease plasma homocysteine levels significantly [96]. On the other hand, no clinical end-point was pursued in this study. Therefore, the observed association between vitamin supplementation and improved health was used as a rationale for conducting many RCTs. That is, evidence first accumulated through observational and retrospective studies, and then this evidence fueled further studies in the form of prospective cohorts and interventional studies (RCTs). This happened with Vitamins A-C-E in 1990s, and then with B group Vitamins in 2000s, and with Vitamin D around and after 2010 (Fig. 2.). On the other hand, most RCTs did not find any evidence supporting the use of vitamin supplementations by general public, supporting the evidence from observational studies that vitamin supplementation is more common among people who are leading a health conscious behavior and diet rich in fruits and vegetables, follow Mediterranean diet; and the observed improvement in those who take vitamin supplementation is probably confounded by this healthy cluster of behavior [97,98]. On the other hand, while studies coming from outside the US/UK generally showed a benefit in these non-RCT studies [69,74], those carried out in the US/UK, using the data reported by those living in the US/UK did not show this improvement [77,78]. Similarly, a study conducted in Sweden found improved outcomes only in a subgroup analysis of people who reported inadequate dietary intake [26]. It is a known fact that adequate dietary vitamin intake is necessary for optimum cardiovascular outcomes. Taking this into account, a difference in dietary habits and nutritional intake from foods could be a reason explaining this difference, since majority of studies conducted in Iran and rural South Korea, where access to adequate nutritious food may be limited, reported improved outcomes after vitamin supplementation. Furthermore, while in the early years Vitamin E, C, and B received the most attention; especially after around 2010 when these observed benefits were mostly debunked by RCTs, the attention has shifted to Vitamin D supplementation (Fig. 2), which went through a similar process and has recently been proven to be futile [75,77,78,80].

Many randomized clinical trials have been conducted repeatedly in different subsets of populations on the basis that non-RCT studies suggested a potential benefit between vitamin supplementation and clinical cardiovascular outcomes. Even though creation and testing of a hypothesis from previously available cohorts is relatively easy and could be a lucrative way to publish, it is important to remember that creating a hypothesis and testing it in observational studies or other non-randomized studies are susceptible to inherent confounding factors, don't necessarily equate to causation, and statistical adjustments are not immune to confounding [99]. Furthermore, it would be plausible to argue that non-RCT studies with negative results are less likely to be submitted for publication (publication bias). Future studies with vitamin supplementation intervention should aim specific populations (such as specific single nucleotide polymorphisms in folate metabolism, those with insufficient dietary intake, or those with a proven low-levels of such vitamin) rather than the general public.

5. Conclusion

This comprehensive review article found no significant evidence between vitamin supplementation and improved clinical cardiovascular outcomes in general public. Therefore, a recommendation to suggest vitamin use to maintain and/or improve clinical cardiovascular outcomes cannot not be made for the general public. Instead; counseling people to follow a healthy-diet, rich in fruits and vegetables seems more appropriate.

Funding

This research did not receive any specific grant from funding agencies in the public, commercial, or not-for-profit sectors.

Author contributions

Conceptualization; BS-UC. Data curation; BS-AS. Formal analysis; BS-AS. Investigation; BS-BHE-AS. Methodology; BS-BHE-AS. Project administration; BS-UC. Supervision; BS-UC. Validation; BS-BHE-AS. Visualization; BS-AS. Roles/Writing; original draft; BS. Writing - review & editing; BS-AS-BHE-UC.

Declaration of competing interest

None declared.

Acknowledgements

None.

References

[1] Global, regional, and national age-sex-specific mortality for 282 causes of death in 195 countries and territories, 1980-2017: a systematic analysis for the Global Burden of Disease Study 2017. Lancet (London, England) 2018;392:1736–88.
[2] Kantor ED, Rehm CD, Du M, White E, Giovannucci EL. Trends in dietary supplement use among US adults from 1999-2012. J Am Med Assoc 2016;316:1464–74.
[3] Guallar E, Stranges S, Mulrow C, Appel LJ, Miller 3rd ER. Enough is enough: stop wasting money on vitamin and mineral supplements. Ann Intern Med 2013;159:850–1.
[4] Manson JE, Bassuk SS. Vitamin and mineral supplements: what clinicians need to know. J Am Med Assoc 2018;319:859–60.
[5] Gillilan RE, Mondell B, Warbasse JR. Quantitative evaluation of vitamin E in the treatment of angina pectoris. Am Heart J 1977;93:444–9.
[6] Enstrom JE, Pauling L. Mortality among health-conscious elderly Californians. Proceedings of the National Academy of Sciences of the United States of America 1982;79:6023–7.
[7] Enstrom JE, Kanim LE, Breslow L. The relationship between vitamin C intake, general health practices, and mortality in Alameda County, California. Am J Public Health 1986;76:1124–30.
[8] Enstrom JE, Kanim LE, Klein MA. Vitamin C intake and mortality among a sample of the United States population. Epidemiology 1992;3:194–202.
[9] DeMaio SJ, King 3rd SB, Lembo NJ, Roubin GS, Hearn JA, Bhagavan HN, et al. Vitamin E supplementation, plasma lipids and incidence of restenosis after percutaneous transluminal coronary angioplasty (PTCA). J Am Coll Nutr 1992;11:68–73.
[10] Stampfer MJ, Hennekens CH, Manson JE, Colditz GA, Rosner B, Willett WC. Vitamin E consumption and the risk of coronary disease in women. N Engl J Med 1993;328:1444–9.
[11] Rimm EB, Stampfer MJ, Ascherio A, Giovannucci E, Colditz GA, Willett WC. Vitamin E consumption and the risk of coronary heart disease in men. N Engl J Med 1993;328:1450–6.
[12] Kushi LH, Folsom AR, Prineas RJ, Mink PJ, Wu Y, Bostick RM. Dietary antioxidant vitamins and death from coronary heart disease in postmenopausal women. N Engl J Med 1996;334:1156–62.
[13] Houston DK, Johnson MA, Daniel TD, Poon LW. Health and dietary characteristics of supplement users in an elderly population. International journal for vitamin and nutrition research Internationale Zeitschrift fur Vitamin- und Ernahrungsforschung Journal international de vitaminologie et de nutrition 1997;67:183–91.
[14] Mayer-Davis EJ, Monaco JH, Marshall JA, Rushing J, Juhaeri. Vitamin C intake and cardiovascular disease risk factors in persons with non-insulin-

dependent diabetes mellitus. From the insulin resistance atherosclerosis study and the san luis valley diabetes study. Prev Med 1997;26:277–83.
[15] Virtamo J, Rapola JM, Ripatti S, Heinonen OP, Taylor PR, Albanes D, et al. Effect of vitamin E and beta carotene on the incidence of primary nonfatal myocardial infarction and fatal coronary heart disease. Arch Intern Med 1998;158:668–75.
[16] Ascherio A, Rimm EB, Hernán MA, Giovannucci E, Kawachi I, Stampfer MJ, et al. Relation of consumption of vitamin E, vitamin C, and carotenoids to risk for stroke among men in the United States. Ann Intern Med 1999;130:963–70.
[17] Lee IM, Cook NR, Manson JE, Buring JE, Hennekens CH. Beta-carotene supplementation and incidence of cancer and cardiovascular disease: the Women's Health Study. Journal of the National Cancer Institute 1999;91:2102–6.
[18] Watkins ML, Erickson JD, Thun MJ, Mulinare J, Heath Jr CW. Multivitamin use and mortality in a large prospective study. American journal of epidemiology 2000;152:149–62.
[19] Salonen RM, Nyyssönen K, Kaikkonen J, Porkkala-Sarataho E, Voutilainen S, Rissanen TH, et al. Six-year effect of combined vitamin C and E supplementation on atherosclerotic progression: the antioxidant supplementation in atherosclerosis prevention (ASAP) study. Circulation 2003;107:947–53.
[20] Stephens NG, Parsons A, Schofield PM, Kelly F, Cheeseman K, Mitchinson MJ. Randomised controlled trial of vitamin E in patients with coronary disease: cambridge Heart Antioxidant Study (CHAOS). Lancet (London, England) 1996;347:781–6.
[21] Rasool AH, Rehman A, Wan Yusuf WN, Rahman AR. Vitamin E and its effect on arterial stiffness in postmenopausal women–a randomized controlled trial. International journal of clinical pharmacology and therapeutics 2003;41:587–92.
[22] Lee DH, Folsom AR, Harnack L, Halliwell B, Jacobs Jr DR. Does supplemental vitamin C increase cardiovascular disease risk in women with diabetes? The American journal of clinical nutrition 2004;80:1194–200.
[23] Hercberg S, Galan P, Preziosi P, Bertrais S, Mennen L, Malvy D, et al. The SU.VI.MAX Study: a randomized, placebo-controlled trial of the health effects of antioxidant vitamins and minerals. Arch Intern Med 2004;164:2335–42.
[24] Lonn E, Bosch J, Yusuf S, Sheridan P, Pogue J, Arnold JM, et al. Effects of long-term vitamin E supplementation on cardiovascular events and cancer: a randomized controlled trial. J Am Med Assoc 2005;293:1338–47.
[25] Lee IM, Cook NR, Gaziano JM, Gordon D, Ridker PM, Manson JE, et al. Vitamin E in the primary prevention of cardiovascular disease and cancer: the Women's Health Study: a randomized controlled trial. J Am Med Assoc 2005;294:56–65.
[26] Messerer M, Håkansson N, Wolk A, Akesson A. Dietary supplement use and mortality in a cohort of Swedish men. Br J Nutr 2008;99:626–31.
[27] Sesso HD, Buring JE, Christen WG, Kurth T, Belanger C, MacFadyen J, et al. Vitamins E and C in the prevention of cardiovascular disease in men: the Physicians' Health Study II randomized controlled trial. J Am Med Assoc 2008;300:2123–33.
[28] Stepaniak U, Micek A, Grosso G, Stefler D, Topor-Madry R, Kubinova R, et al. Antioxidant vitamin intake and mortality in three Central and Eastern European urban populations: the HAPIEE study. Eur J Nutr 2016;55:547–60.
[29] Rimm EB, Willett WC, Hu FB, Sampson L, Colditz GA, Manson JE, et al. Folate and vitamin B6 from diet and supplements in relation to risk of coronary heart disease among women. J Am Med Assoc 1998;279:359–64.
[30] Merchant AT, Hu FB, Spiegelman D, Willett WC, Rimm EB, Ascherio A. The use of B vitamin supplements and peripheral arterial disease risk in men are inversely related. J Nutr 2003;133:2863–7.
[31] He K, Merchant A, Rimm EB, Rosner BA, Stampfer MJ, Willett WC, et al. Folate, vitamin B6, and B12 intakes in relation to risk of stroke among men. Stroke 2004;35:169–74.
[32] Toole JF, Malinow MR, Chambless LE, Spence JD, Pettigrew LC, Howard VJ, et al. Lowering homocysteine in patients with ischemic stroke to prevent recurrent stroke, myocardial infarction, and death: the Vitamin Intervention for Stroke Prevention (VISP) randomized controlled trial. J Am Med Assoc 2004;291:565–75.
[33] Andersson SE, Edvinsson ML, Edvinsson L. Reduction of homocysteine in elderly with heart failure improved vascular function and blood pressure control but did not affect inflammatory activity. Basic Clin Pharmacol Toxicol 2005;97:306–10.
[34] Lonn E, Yusuf S, Arnold MJ, Sheridan P, Pogue J, Micks M, et al. Homocysteine lowering with folic acid and B vitamins in vascular disease. N Engl J Med 2006;354:1567–77.
[35] Saposnik G, Ray JG, Sheridan P, McQueen M, Lonn E. Homocysteine-lowering therapy and stroke risk, severity, and disability: additional findings from the HOPE 2 trial. Stroke 2009;40:1365–72.
[36] Zoungas S, McGrath BP, Branley P, Kerr PG, Muske C, Wolfe R, et al. Cardiovascular morbidity and mortality in the Atherosclerosis and Folic Acid Supplementation Trial (ASFAST) in chronic renal failure: a multicenter, randomized, controlled trial. J Am Coll Cardiol 2006;47:1108–16.
[37] Jamison RL, Hartigan P, Kaufman JS, Goldfarb DS, Warren SR, Guarino PD, et al. Effect of homocysteine lowering on mortality and vascular disease in advanced chronic kidney disease and end-stage renal disease: a randomized controlled trial. J Am Med Assoc 2007;298:1163–70.
[38] Ishihara J, Iso H, Inoue M, Iwasaki M, Okada K, Kita Y, et al. Intake of folate, vitamin B6 and vitamin B12 and the risk of CHD: the Japan public health center-based prospective study cohort I. J Am Coll Nutr 2008;27:127–36.
[39] Albert CM, Cook NR, Gaziano JM, Zaharris E, MacFadyen J, Danielson E, et al. Effect of folic acid and B vitamins on risk of cardiovascular events and total mortality among women at high risk for cardiovascular disease: a randomized trial. J Am Med Assoc 2008;299:2027–36.
[40] Galan P, de Bree A, Mennen L, Potier de Courcy G, Preziozi P, Bertrais S, et al. Background and rationale of the SU.FOL.OM3 study: double-blind randomized placebo-controlled secondary prevention trial to test the impact of supplementation with folate, vitamin B6 and B12 and/or omega-3 fatty acids on the prevention of recurrent ischemic events in subjects with atherosclerosis in the coronary or cerebral arteries. J Nutr Health Aging 2003;7:428–35.
[41] Galan P, Briancon S, Blacher J, Czernichow S, Hercberg S. The SU.FOL.OM3 Study: a secondary prevention trial testing the impact of supplementation with folate and B-vitamins and/or Omega-3 PUFA on fatal and non fatal cardiovascular events, design, methods and participants characteristics. Trials 2008;9:35.
[42] Galan P, Kesse-Guyot E, Czernichow S, Briancon S, Blacher J, Hercberg S. Effects of B vitamins and omega 3 fatty acids on cardiovascular diseases: a randomised placebo controlled trial. BMJ (Clinical research ed) 2010;341:c6273.
[43] Szabo de Edelenyi F, Vergnaud AC, Ahluwalia N, Julia C, Hercberg S, Blacher J, et al. Effect of B-vitamins and n-3 PUFA supplementation for 5 years on blood pressure in patients with CVD. Br J Nutr 2012;107:921–7.
[44] Armitage JM, Bowman L, Clarke RJ, Wallendszus K, Bulbulia R, Rahimi K, et al. Effects of homocysteine-lowering with folic acid plus vitamin B12 vs placebo on mortality and major morbidity in myocardial infarction survivors: a randomized trial. J Am Med Assoc 2010;303:2486–94.
[45] Løland KH, Bleie O, Blix AJ, Strand E, Ueland PM, Refsum H, et al. Effect of homocysteine-lowering B vitamin treatment on angiographic progression of coronary artery disease: a Western Norway B Vitamin Intervention Trial (WENBIT) substudy. Am J Cardiol 2010;105:1577–84.
[46] Kwok T, Chook P, Qiao M, Tam L, Poon YK, Ahuja AT, et al. Vitamin B-12 supplementation improves arterial function in vegetarians with subnormal vitamin B-12 status. J Nutr Health Aging 2012;16:569–73.
[47] Peña AS, Maftei O, Dowling K, Gent R, Wiltshire E, MacKenzie K, et al. Folate fortification and supplementation do not provide vascular health benefits in type 1 diabetes. J Pediatr 2013;163:255–60.
[48] van Dijk SC, Enneman AW, Swart KM, van Wijngaarden JP, Ham AC, Brouwer-Brolsma EM, et al. Effects of 2-year vitamin B12 and folic acid supplementation in hyperhomocysteinemic elderly on arterial stiffness and cardiovascular outcomes within the B-PROOF trial. J Hypertens 2015;33:1897–906. discussion 906.
[49] Hsia J, Heiss G, Ren H, Allison M, Dolan NC, Greenland P, et al. Calcium/vitamin D supplementation and cardiovascular events. Circulation 2007;115:846–54.
[50] Prentice RL, Pettinger MB, Jackson RD, Wactawski-Wende J, Lacroix AZ, Anderson GL, et al. Health risks and benefits from calcium and vitamin D supplementation: women's Health Initiative clinical trial and cohort study. Osteoporos Int : a journal established as result of cooperation between the European Foundation for Osteoporosis and the National Osteoporosis Foundation of the USA 2013;24:567–80.
[51] Donneyong MM, Hornung CA, Taylor KC, Baumgartner RN, Myers JA, Eaton CB, et al. Risk of heart failure among postmenopausal women: a secondary analysis of the randomized trial of vitamin D plus calcium of the women's health initiative. Circulation Heart failure 2015;8:49–56.
[52] Wang L, Manson JE, Buring JE, Lee IM, Sesso HD. Dietary intake of dairy products, calcium, and vitamin D and the risk of hypertension in middle-aged and older women. Hypertension (Dallas, Tex : 1979) 2008;51:1073–9.
[53] Witham MD, Dove FJ, Sugden JA, Doney AS, Struthers AD. The effect of vitamin D replacement on markers of vascular health in stroke patients - a randomised controlled trial. Nutr Metabol Cardiovasc Dis : Nutr Metabol Cardiovasc Dis 2012;22:864–70.
[54] Messenger W, Nielson CM, Li H, Beer T, Barrett-Connor E, Stone K, et al. Serum and dietary vitamin D and cardiovascular disease risk in elderly men: a prospective cohort study. Nutr Metabol Cardiovasc Dis : Nutr Metabol Cardiovasc Dis 2012;22:856–63.
[55] Bajaj A, Stone KL, Peters K, Parimi N, Barrett-Connor E, Bauer D, et al. Circulating vitamin D, supplement use, and cardiovascular disease risk: the MrOS Sleep Study. The Journal of clinical endocrinology and metabolism 2014;99:3256–62.
[56] Gepner AD, Ramamurthy R, Krueger DC, Korcarz CE, Binkley N, Stein JH. A prospective randomized controlled trial of the effects of vitamin D supplementation on cardiovascular disease risk. PloS One 2012;7:e36617.
[57] Sokol SI, Srinivas V, Crandall JP, Kim M, Tellides G, Lebastchi AH, et al. The effects of vitamin D repletion on endothelial function and inflammation in patients with coronary artery disease. Vasc Med 2012;17:394–404.
[58] Witham MD, Adams F, Kabir G, Kennedy G, Belch JJ, Khan F. Effect of short-term vitamin D supplementation on markers of vascular health in South Asian women living in the UK–a randomised controlled trial. Atherosclerosis 2013;230:293–9.
[59] Witham MD, Adams F, McSwiggan S, Kennedy G, Kabir G, Belch JJ, et al. Effect of intermittent vitamin D3 on vascular function and symptoms in chronic fatigue syndrome–a randomised controlled trial. Nutr Metabol Cardiovasc Dis : Nutr Metabol Cardiovasc Dis 2015;25:287–94.
[60] Arora P, Song Y, Dusek J, Plotnikoff G, Sabatine MS, Cheng S, et al. Vitamin D therapy in individuals with prehypertension or hypertension: the DAYLIGHT trial. Circulation 2015;131:254–62.

[61] Pilz S, Gaksch M, Kienreich K, Grübler M, Verheyen N, Fahrleitner-Pammer A, et al. Effects of vitamin D on blood pressure and cardiovascular risk factors: a randomized controlled trial. Hypertension (Dallas, Tex : 1979) 2015;65:1195–201.
[62] Dong JY, Iso H, Kitamura A, Tamakoshi A. Multivitamin use and risk of stroke mortality: the Japan collaborative cohort study. Stroke 2015;46:1167–72.
[63] Robbins J, Petrone AB, Gaziano JM, Djoussé L. Dietary vitamin D and risk of heart failure in the physicians' health study. Clinical nutrition (Edinburgh, Scotland) 2016;35:650–3.
[64] Gepner AD, Haller IV, Krueger DC, Korcarz CE, Binkley N, Stein JH. A randomized controlled trial of the effects of vitamin D supplementation on arterial stiffness and aortic blood pressure in Native American women. Atherosclerosis 2015;240:526–8.
[65] Thiele I, Linseisen J, Meisinger C, Schwab S, Huth C, Peters A, et al. Associations between calcium and vitamin D supplement use as well as their serum concentrations and subclinical cardiovascular disease phenotypes. Atherosclerosis 2015;241:743–51.
[66] Bolland MJ, Grey A, Gamble GD, Reid IR. Concordance of results from randomized and observational analyses within the same study: a Re-analysis of the women's health initiative limited-access dataset. PloS One 2015;10:e0139975.
[67] Miskulin DC, Majchrzak K, Tighiouart H, Muther RS, Kapoian T, Johnson DS, et al. Ergocalciferol supplementation in hemodialysis patients with vitamin D deficiency: a randomized clinical trial. J Am Soc Nephrol : JASN (J Am Soc Nephrol) 2016;27:1801–10.
[68] Harris E, Rowsell R, Pipingas A, Macpherson H. No effect of multivitamin supplementation on central blood pressure in healthy older people: a randomized controlled trial. Atherosclerosis 2016;246:236–42.
[69] Kang JY, Kim MK, Jung S, Shin J, Choi BY. The cross-sectional relationships of dietary and serum vitamin D with cardiometabolic risk factors: metabolic components, subclinical atherosclerosis, and arterial stiffness. Nutrition 2016;32:1048–10456. e1.
[70] Salekzamani S, Bavil AS, Mehralizadeh H, Jafarabadi MA, Ghezel A, Gargari BP. The effects of vitamin D supplementation on proatherogenic inflammatory markers and carotid intima media thickness in subjects with metabolic syndrome: a randomized double-blind placebo-controlled clinical trial. Endocrine 2017;57:51–9.
[71] Scragg R, Stewart AW, Waayer D, Lawes CMM, Toop L, Sluyter J, et al. Effect of monthly high-dose vitamin D supplementation on cardiovascular disease in the vitamin D assessment study : a randomized clinical trial. JAMA cardiology 2017;2:608–16.
[72] Kumar V, Yadav AK, Lal A, Kumar V, Singhal M, Billot L, et al. A randomized trial of vitamin D supplementation on vascular function in CKD. J Am Soc Nephrol : JASN (J Am Soc Nephrol) 2017;28:3100–8.
[73] Kendrick J, Andrews E, You Z, Moreau K, Nowak KL, Farmer-Bailey H, et al. Cholecalciferol, calcitriol, and vascular function in CKD: a randomized, double-blind trial. Clin J Am Soc Nephrol : CJASN 2017;12:1438–46.
[74] Khayyatzadeh SS, Mirmoosavi SJ, Fazeli M, Abasalti Z, Avan A, Javandoost A, et al. High-dose vitamin D supplementation is associated with an improvement in several cardio-metabolic risk factors in adolescent girls: a nine-week follow-up study. Ann Clin Biochem 2018;55:227–35.
[75] Tomson J, Hin H, Emberson J, Kurien R, Lay M, Cox J, et al. Effects of vitamin D on blood pressure, arterial stiffness, and cardiac function in older people after 1 Year: BEST-D (biochemical efficacy and safety trial of vitamin D). Journal of the American Heart Association 2017;6.
[76] Sluyter JD, Camargo Jr CA, Stewart AW, Waayer D, Lawes CMM, Toop L, et al. Effect of monthly, high-dose, long-term vitamin D supplementation on central blood pressure parameters: a randomized controlled trial substudy. Journal of the American Heart Association 2017;6.
[77] Harvey NC, D'Angelo S, Paccou J, Curtis EM, Edwards M, Raisi-Estabragh Z, et al. Calcium and vitamin D supplementation are not associated with risk of incident ischemic cardiac events or death: findings from the UK Biobank cohort. J Bone Miner Res : the official journal of the American Society for Bone and Mineral Research 2018;33:803–11.
[78] Manson JE, Cook NR, Lee IM, Christen W, Bassuk SS, Mora S, et al. Vitamin D supplements and prevention of cancer and cardiovascular disease. N Engl J Med 2019;380:33–44.
[79] Boursiquot BC, Larson JC, Shalash OA, Vitolins MZ, Soliman EZ, Perez MV. Vitamin D with calcium supplementation and risk of atrial fibrillation in postmenopausal women. Am Heart J 2019;209:68–78.
[80] Rajakumar K, Moore CG, Khalid AT, Vallejo AN, Virji MA, Holick MF, et al. Effect of vitamin D3 supplementation on vascular and metabolic health of vitamin D-deficient overweight and obese children: a randomized clinical trial. The American journal of clinical nutrition 2020;111:757–68.
[81] Wang C, Li Y, Zhu K, Dong YM, Sun CH. Effects of supplementation with multivitamin and mineral on blood pressure and C-reactive protein in obese Chinese women with increased cardiovascular disease risk. Asia Pac J Clin Nutr 2009;18:121–30.
[82] Neuhouser ML, Wassertheil-Smoller S, Thomson C, Aragaki A, Anderson GL, Manson JE, et al. Multivitamin use and risk of cancer and cardiovascular disease in the Women's Health Initiative cohorts. Arch Intern Med 2009;169:294–304.
[83] Hara A, Sasazuki S, Inoue M, Shimazu T, Iwasaki M, Sawada N, et al. Use of vitamin supplements and risk of total cancer and cardiovascular disease among the Japanese general population: a population-based survey. BMC Publ Health 2011;11:540.
[84] Li K, Kaaks R, Linseisen J, Rohrmann S. Vitamin/mineral supplementation and cancer, cardiovascular, and all-cause mortality in a German prospective cohort (EPIC-Heidelberg). Eur J Nutr 2012;51:407–13.
[85] Lamas GA, Boineau R, Goertz C, Mark DB, Rosenberg Y, Stylianou M, et al. Oral high-dose multivitamins and minerals after myocardial infarction: a randomized trial. Ann Intern Med 2013;159:797–805.
[86] Rautiainen S, Lee IM, Rist PM, Gaziano JM, Manson JE, Buring JE, et al. Multivitamin use and cardiovascular disease in a prospective study of women. The American journal of clinical nutrition 2015;101:144–52.
[87] Bailey RL, Fakhouri TH, Park Y, Dwyer JT, Thomas PR, Gahche JJ, et al. Multivitamin-mineral use is associated with reduced risk of cardiovascular disease mortality among women in the United States. J Nutr 2015;145:572–8.
[88] Vučković BA, van Rein N, Cannegieter SC, Rosendaal FR, Lijfering WM. Vitamin supplementation on the risk of venous thrombosis: results from the MEGA case-control study. The American journal of clinical nutrition 2015;101:606–12.
[89] Fulton RL, McMurdo ME, Hill A, Abboud RJ, Arnold GP, Struthers AD, et al. Effect of vitamin K on vascular health and physical function in older people with vascular disease–A randomised controlled trial. J Nutr Health Aging 2016;20:325–33.
[90] Rautiainen S, Rist PM, Glynn RJ, Buring JE, Gaziano JM, Sesso HD. Multivitamin use and the risk of cardiovascular disease in men. J Nutr 2016;146:1235–40.
[91] Rautiainen S, Wang L, Lee IM, Manson JE, Gaziano JM, Buring JE, et al. Multivitamin use and the risk of hypertension in a prospective cohort study of women. J Hypertens 2016;34:1513–9.
[92] Adebamowo SN, Feskanich D, Stampfer M, Rexrode K, Willett WC. Multivitamin use and risk of stroke incidence and mortality amongst women. Eur J Neurol 2017;24:1266–73.
[93] Chen F, Du M, Blumberg JB, Ho Chui KK, Ruan M, Rogers G, et al. Association among dietary supplement use, nutrient intake, and mortality among U.S. Adults: a cohort study. Ann Intern Med 2019;170:604–13.
[94] Gey KF, Brubacher GB, Stähelin HB. Plasma levels of antioxidant vitamins in relation to ischemic heart disease and cancer. The American journal of clinical nutrition 1987;45:1368–77.
[95] Kok FJ, de Bruijn AM, Vermeeren R, Hofman A, van Laar A, de Bruin M, et al. Serum selenium, vitamin antioxidants, and cardiovascular mortality: a 9-year follow-up study in The Netherlands. The American journal of clinical nutrition 1987;45:462–8.
[96] Ward M, McNulty H, McPartlin J, Strain JJ, Weir DG, Scott JM. Plasma homocysteine, a risk factor for cardiovascular disease, is lowered by physiological doses of folic acid. QJM : monthly journal of the Association of Physicians 1997;90:519–24.
[97] Hernández-Díaz S, Martínez-Losa E, Fernández-Jarne E, Serrano-Martínez M, Martínez-González MA. Dietary folate and the risk of nonfatal myocardial infarction. Epidemiology 2002;13:700–6.
[98] Hu FB, Willett WC. Optimal diets for prevention of coronary heart disease. J Am Med Assoc 2002;288:2569–78.
[99] Haslam A, Prasad V. Multivitamins do not reduce cardiovascular disease and mortality and should not Be taken for this purpose: how do we know that? Circulation Cardiovascular quality and outcomes 2018;11:e004886.