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Review

Vascular action of polyphenols

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Dietary patterns are widely recognised as contributors to cardiovascular and cerebrovascular disease. Endothelial function, the elastic properties of large arteries and the magnitude and timing of wave reflections are important determinants of cardiovascular performance. Several epidemiological studies suggest that the regular consumption of foods and beverages rich in flavonoids is associated with a reduction in the risk of several pathological conditions ranging from hypertension to coronary heart disease, stroke and dementia. The impairment of endothelial function is directly related to ageing and an association between decreased cerebral perfusion and dementia has been shown to exist. Cerebral blood flow (CBF) must be maintained to ensure a constant delivery of oxygen and glucose as well as the removal of waste products. Increasing blood flow is one potential way for improving brain function and the prospect for increasing CBF with dietary polyphenols is extremely promising. The major polyphenols shown to have some of these effects in humans are primarily from cocoa, wine, grape seed, berries, tea, tomatoes (polyphenolics and nonpolyphenolics), soy and pomegranate. There has been a significant paradigm shift in polyphenol research during the last decade. This review summarises our current knowledge in this area and points the way for the development of new types of functional foods targeted to brain health through improving vascular health.

Keywords: Blood flow / Cognition / Heart / Polyphenols / Vascular disorder

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1 Introduction

Several epidemiological studies suggest that regular consumption of foods and beverages rich in polyphenols is associated with a reduction in the risk of a range of pathological conditions, ranging from hypertension to coronary heart disease (CHD), stroke and dementia [1 – 5]. Endothelial function, elastic properties of large arteries, and the magnitude and timing of wave reflections are important determinants of cardiovascular performance. The prevention of 'life-style related atherothrombotic diseases' which can result in cerebral infarction and stroke is an important and urgent social issue in many developed countries, and especially concern the ageing 'baby boomer' population. Epidemiologic studies have provided strong evidence for a causative role of an inappropriate diet in the development and clinical outcome of thrombotic disease. Marked as the top killers, cardiovascular (CVD) and cerebrovascular (CVA) disease are understandably the target of a myriad of health products promising to help those interested in reducing the risk or severity of these diseases.

The impairment of endothelial function is a nearly universal accompaniment to the ageing process and decreased cerebral perfusion has been recently associated with dementia [3, 6]. Cerebral ischemia results from the reduction or prevention of cerebral blood flow (CBF) and is usually caused by the occlusion of a cerebral artery by a thrombotic clot originating from the peripheral circulatory system, or due to intracerebral haemorrhage. The brain is particularly susceptible to a loss or reduction in blood flow, unlike peripheral organs which can survive several hours of complete ischemia. Thus CBF must be maintained to ensure a constant delivery of oxygen and glucose as well as the removal of waste products. Increasing CBF is one potential way of improving brain function and the prospect

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Abbreviations: BCA, blackcurrant anthocyanins; **CAD,** coronary artery disease; **CBF,** cerebral blood flow; **CFR,** coronary flow reserve; **CHD,** coronary heart disease; **eNOS,** endothelial nitric oxide synthase; **FMD,** flow-mediated dilation; **GSE,** grape seed extract; **HDL,** high density lipoprotein; **IMTmax,** maxima intima-media thickness; **LDL,** low density lipoprotein; **NO,** nitric oxide; **PJ,** pomegranate juice of achieving this with dietary polyphenols seems extremely promising [1, 6]. Recent reports have provided a link between CBF and brain function, suggesting that an increase in CBF is beneficial for cognitive function [6]. Obesity has also been suggested as a risk factor for ischemia, haemorrhagic stroke and vascular dementia [7, 8]. Obesity is strongly related to higher blood pressure or hypertension, the development of atherosclerotic lesions and to reduced cognitive function [9, 10]. In summary, the main risk factors that lead to the occurrence of stroke are hypertension, atherosclerosis, high circulating levels of low density lipoprotein (LDL)-cholesterol and diabetes. Of note, is the fact that atherosclerotic lesions are most commonly initiated by oxidative damage to blood vessel walls, after which high levels of LDL intake then causes these lesions to enlarge resulting in the formation of fatty deposits on blood vessel walls leading to a restriction of blood flow, complete occlusion or the release of thrombotic or fatty deposits which can then occlude distal blood vessels [11]. Genetic factors are also involved, including polymorphisms in the genes coding for angiotensin converting enzyme (ACE), endothelial nitric oxide synthase (eNOS) and β-fibrinogen [12].

2 Polyphenols

Phenolic compounds are abundant micronutrients in our diet with an average consumption of around 1 g/day [13]. The phenolic content of fruit, vegetables and other plant foods varies considerably, not only between different types but also between cultivars of the same type and can even depend on growing condition and the time of harvest. Fruits and vegetables are a particularly rich source of polyphenols and many polyphenols are well known for their antioxidant activity [14]. Polyphenols are a group of chemical substances, characterised by the presence of more than one phenolic group whereas the phenolic acids are phenols with only one ring. Polyphenols belong to one of the major classes of plant secondary metabolites including, flavonoids, lignans, stilbenes, coumarins and tannins [15]. Several thousand polyphenols have been identified in edible plants and they are divided into different groups according to their structure and complexity [16]. Flavonoids are the largest group of phenolic compounds and have a basic skeleton composed of three rings. They are classified into six families according to their substitution pattern, and include anthocyanins, flavones, isoflavones, flavonols, flavanones and flavanols. Even though the term 'polyphenol' encompasses more than 8000 different structures, only a limited number have been studied pharmacologically. For example, red wine contains the natural phytoalexin resveratrol and the flavonoids quercetin, delphinidin and (+)-catechin, which have been well studied and shown to possess the pharmacological properties explaining the beneficial effects of moderate red wine consumption against the onset of CVD and CVA [17–19].

The major phytochemicals (primarily polyphenols) which have been shown to have cerebrovascular effects are primarily from cocoa, wine, grape seed, berries, tea, tomatoes, soy and pomegranate (Table 1). A systematic search was undertaken of the following scientific databases – Medline (1996 to present), PubMed (1996 to present) and Science Direct (1996 to present). We mostly considered human clinical trial data, but also included animal experiments where human data were not available. The purpose of this review is to discuss the roles of fruits and vegetables in relation to the aetiology and prevention of cerebrovascular disease.

3 Grape seed extract (GSE) and wine

One of the best-known sources of flavones is GSE. A research review noted that the flavonoids within grape products may improve endothelial function and offer increased protection against LDL oxidation [20]. A commercially sponsored study reported high plasma antioxidant activity in subjects taking 600 mg of commercial GSE (MegaNatural Gold from Madera), as well as with the consumption of grapes, grape juice, red wine and other GSEs [21]. Subjects with high cholesterol also showed a change in plasma LDL, high density lipoprotein (HDL) and total cholesterol levels. GSE may also work well in combination with other nutritional therapies. A pilot study demonstrated the ability of GSE (as ActiVin from San Joaquin Valley Concentrates) plus niacin-bound chromium (as Chrome-Mate from InterHealth Nutraceuticals, USA) to reduce cholesterol levels [22]. Researchers gave 40 hypercholesterolemic subjects a daily dose of 200 μg of chromium, 100 mg of GSE, chromium plus GSE or a placebo only for 2 months. They found that the combination of chromium and GSE significantly decreased both total cholesterol and LDL levels. In another human study, Clifton [23] demonstrated that the consumption of 2 g of GSE *per* day could improve flow-mediated dilation (FMD) in healthy adults and suggested that this might be mediated by changes in the production of nitric oxide (NO). The effect of NO was assessed by changes in cell adhesion, blood clotting and changes in the level of fibrinolytic molecules. The dose of grape seed proanthocyanidins extract (GSPE) required to induce these changes however, was equivalent to a daily intake of some 3.5 g for a 70-kg human [24]. Most over-the-counter forms of GSPE however, range in content from 50 to 100 mg with a recommended daily dose of one capsule and are thus unlikely to produce these beneficial effects.

On the basis of epidemiological studies, the moderate intake of alcoholic beverages, including red wine, reduces the risk of cardiovascular, cerebrovascular and peripheral

Table 1. The effects of polyphenols on vascular action: a quality rating assessment [90]

Source of polyphenols	Study design	References	Quantification and outcome	Quality of study	Overall level of evidence
GSE (MegaNatural® Gold)	Diet intervention trial, $n = 9$, 3 men and 6 women, age 25–58 years	[21]	Plasma cholesterol, antioxidant capacity, low-density lipoprotein cholesterol and HDL cholesterol concentrations were significantly decreased in the subjects with high cholesterol	Medium	Possible
GSE in yoghurt	Randomised controlled crossover trial, $n = 36$, 24 men and 12 women	[23]	GSE alone, or with added quercetin in yoghurt, alters vascular function, endothelial function and degree of oxidative damage in comparison to control yoghurt. GSE alone also improved flow-mediated dilatation of blood vessel	High	Possible
Red wine	Observational, prospective cohort study, $n = 1828$, age 18–65 years	[85]	The effects of the consumption of wine, beer and distilled spirits on total mortality and on mortality from cardiovascular disease were investigated and mortality rate was recorded after 22 years. A low to moderate intake of wine seems, unlike the consumption of distilled spirits and beer, to be associated with reduced total mortality and reduced mortality from cardiovascular disease	Medium	Insufficient
Red wine	Randomised controlled crossover trial, $n = 22$ healthy men, age 23 years	[19]	The CFR was determined using transthoracic Doppler echocardiography to measure the effect of red wine and dealcoholised red wine on CFR. An increased CFR was reported with a moderate dose of red wine, but not dealcoholised red wine	Medium	Possible
Black tea	Randomised controlled crossover trial, $n = 66$ with proven CAD	[86]	Both short- and long-term effects of tea were examined. Both interventions improved endothelium-dependent FMD of the brachial artery. Plasma flavonoids also increased after short- and long-term tea consumption	Medium	Possible
Black tea	Randomised controlled parallel study, $n = 21$, 16 healthy men and 5 postmenopausal women	[89]	Endothelium-dependent dilatation of the brachial artery was assessed ultra-sonographically. Regular ingestion of black tea resulted in a significant and consistent increase in endothelium-dependent dilatation compared to control	Medium	Possible
Blackcurrant	Randomised, double-blind, placebo-controlled, crossover study, $n = 9$ healthy men, age 30 years	[42]	Peripheral circulation dynamics either at rest or during typing work was studied. The results suggested that intake of Blackcurrant concentrate may improve shoulder stiffness caused by typing work by increasing peripheral blood flow and reducing muscle fatigue	Low	Insufficient
Cocoa	Randomised, double-blind, placebo-controlled, crossover study, $n = 16$ healthy men, age 25–32 years	[45]	FMD for endothelial dysfunction and reactive hyperemia-induced PAT for microvascular function were used in this study. Collectively, results demonstrated that the human ingestion of flavanol-rich cocoa has at least in part, significant vascular effects	High	Probable
Cocoa	Prospective meta-analysis of 5 randomised controlled studies, involving 173 subjects	[49]	In this meta-analysis, diets rich in cocoa were associated with a statistically significant reduction in both systolic and diastolic blood pressure	High	Probable
Dark chocolate	Randomised controlled crossover trial, $n = 15$ healthy subjects, 7 men and 8 women, age 34 years	[50]	Oral-glucose tolerance tests, blood pressure and blood hematochemical tests were measured. The current study demonstrated that cocoa ingestion not only decreased blood pressure but also improved insulin sensitivity in healthy persons	Medium	Possible
Dark chocolate	Randomised, single-blind, sham procedure-controlled, crossover trial, $n = 17$ healthy individuals, 12 men and 5 women, age 29 years	[51]	FMD of the brachial artery, aortic augmentation index, and carotid–femoral pulse wave velocity were used as measures of endothelial function, wave reflections and aortic stiffness, respectively. Results indicated that dark chocolate acutely dilates muscular arteries, decreases wave reflections and may improve endothelial function in healthy humans	Medium	Possible

Table 1. Continued

Source of polyphenols	Study design	References	Quantification and outcome	Quality of study	Overall level of evidence
Pomegranate	Randomised, placebo-controlled trial, $n = 19$ subjects with vascular disorders, 14 men and 5 women, age 65–75 years	[80]	Common carotid artery IMT, atherosclerotic plaques at the common carotid arteries and the carotid bulb and flow velocities were measured. The results suggested that pomegranate juice consumption by patients with carotid artery stenosis decreases carotid IMT and systolic blood pressure	Medium	Possible
Lycopene (nonpolyphenolics) from tomato	Relationship between plasma levels of lipid-soluble lycopene, carotid maxima intima-media thickness (IMTmax) and several adhesion molecules. Study included 11 healthy control subjects, 11 patients with uncomplicated hypertension, and 11 patients with hypertension and vascular diseases. Age-around 57 years	[62]	IMTmax, an index of atherosclerotic extension/severity, and several soluble adhesion molecules were measured. A statistically significant correlation was found between lycopene and IMTmax, but not with soluble adhesion molecules tested. Overall, results indicated a protective role of this natural dietary antioxidant in atherosclerosis	Medium	Insufficient
Lycopene with and without olive oil	Randomised controlled crossover trial, $n = 21$ healthy subjects, age 22–70 years	[88]	Serum lycopene concentration, total cholesterol, LDL cholesterol, HDL cholesterol, and triacylglycerols were measured. Results demonstrated that the diet high in olive oil and rich in lycopene may decrease the risk of CHD by improving the serum lipid profile compared with a high-carbohydrate, low-fat, lycopene-rich diet	Medium	Insufficient

vascular disease in humans [19, 25]. A meta-analysis of 35 observational studies (including 19 cohort studies), reported that light to moderate red wine consumption may be protective against total and ischemic stroke [26]. This effect may be related to the antithrombotic effect red wine has been shown to have in various experimental thrombotic models. A special red wine (Chateauneuf-du-Pape 1987) which was brewed from a particular type of grape and a grape juice (Welch's natural purple grape juice) exerted a significant antithrombotic effect [27]. A recent randomised controlled crossover human study with a moderate dose of red wine (not dealcoholised) demonstrated an improvement of coronary flow reserve (CFR) [19]. The authors also advocated that the increase of CFR is probably mediated by mechanisms other than the direct antioxidant activity of polyphenols.

Red wine polyphenols have also been shown to regulate eNOS expression and synthesis. Increased active eNOS levels may prevent the development of atherosclerosis *via* its vasoprotective and vasodilatory effects [28]. Therapeutic angiogenesis, which improves blood flow to ischemic tissue by the induction of neovascularisation *via* angiogenic agents, has recently emerged as a promising investigational strategy for the treatment of patients with ischemic limb or heart disease. The polyphenol 3,4′,5-transhydroxy-trans-stilbene (a nonflavonoid), better known as resveratrol, has been purported to have many health benefits including cerebrovascular protective effects. Following two pioneering publications in the field [29, 30], more than 900 original research articles have been published to support the beneficial effect of resveratrol. The approach of using resveratrol (mainly from grapes and berries) to induce angiogenesis and to increase the expression of growth factors and their receptors is an exciting and potentially important strategy for myocardial protection [31]. The heart and the brain are two unique examples of tissues with weak defences, as evidenced by infarct damage following ischemia/reperfusion. Resveratrol has been shown to have a unique effect on neuronal cell death and inflammatory processes, which are important therapeutic targets in the development of both acute and/or chronic neurodegenerative diseases [32, 33]. The dynamic interactions between cells belonging to the 'neurovascular unit', including endothelial cells, astrocytes, pericytes and neurones is considered to be a very important factor in the aetiology of neurodegenerative disease and during the recovery from cerebral stroke [1]. Moreover, inflammation also plays an important role in injury of the neurovascular unit during stroke [1]. Neuronal and glial cell death as a consequence of ischemic injury to the brain or spinal cord largely occurs during the reperfusion stage when cytotoxic factors flood the affected areas. The NO-enhancing and antioxidative properties of resveratrol have been shown to specifically reduce cell loss during the reperfusion stage in experimental injury models using both rabbits [33] and pigs [34]. Although there remains some intense controversy regarding the dose and study design [35], all the trials showed some cardio- or neuro-protective properties of resveratrol. In summary, it appears that resveratrol can act through a variety of mechanisms to impart its protective effect including the specific up regulation of protective genes as well as its more global antioxidative effects [36].

4 Berries

Berries are a rich source of polyphenols, in particular anthocyanins and flavonols. Anthocyanins are water-soluble glycosides of anthocyanidins, polyhydroxyl and polymethoxyl derivatives of 2-phenylbenzopyrylium or flavylium salts. The six anthocyanidins most commonly found in plants are classified according to the number and position of hydroxyl groups on the flavan nucleus, and are named cyanidin (cy), delphinidin (dp), malvidin (mv), peonidin (pn), pelargonidin (pg) and petunidin (pt). Anthocyanins are rapidly absorbed from the digestive tract into plasma after oral administration but do not accumulate in plasma [37, 38]. Although anthocyanins are taken up by brain endothelial cell lines and can possibly cross the monolayer in blood–brain barrier models *in vitro* [39], this has not been verified *in vivo*. Very recently, Shin *et al.* [40] demonstrated that pretreatment with high dose oral anthocyanins (300 mg/kg) could prevent some neuronal cell death in a rat model of ischemic brain injury. Although neuroprotective substances that require pretreatment are of little therapeutic use, this result does highlight the possible benefits of a high anthocyanin diet in the prevention of, or minimisation of the damage caused by, ischemic disease.

Blackcurrant anthocyanins (BCA, 50 mg in a cup of juice) have been shown to be significantly efficacious at reducing visual fatigue by increasing the blood flow in peripheral vessels, and relaxing ciliary smooth muscle tension (Nakaishi, H., Cassis University, http://www.cassis-univ.jp, 2005). In a double blind randomised placebo-controlled, crossover study using healthy female subjects, a significant improvement of skin blood circulation was observed after ingesting 140 mg blackcurrant polyphenols (50 mg anthocyanins) [41]. This improvement in circulation was accompanied by a significant difference in the melanin index, erythema index and skin brightness of the lower eyelid. Another human study investigated the effect of BCA intake (17 mg/kg) on peripheral circulation during rest and typing work using near-infrared spectroscopy (NIRS), and also assessed any improvement in shoulder stiffness mediated by poor local circulation [42]. The results suggested that intake of BCA may improve shoulder stiffness caused by typing work, by increasing peripheral blood flow and thereby reducing muscle fatigue.

5 Cocoa and dark chocolate

The short- and long-term ingestion of cocoa and dark chocolate, particularly rich in flavanols (a subclass of flavonoids), has been shown to induce a consistent and striking peripheral vasodilation in healthy people [3, 4]. The flavanols present in cocoa and chocolate include the monomers, (–)-epicatechin and (+)-catechin as well as the oligomers of these monomeric units, procyanidins. In a recent long-term (12 wk) intervention study in women (18–65 years), the intake of a high flavanol (329 mg) cocoa drink was associated with a significant increase in blood flow in cutaneous and subcutaneous tissue, as well as an improved resistance against UV-induced erythema [43]. This increase in cutaneous blood flow associated with cocoa intake is consistent with other human trials which report enhanced FMD of conduit arteries and augmented microcirculation after the ingestion of flavanol-rich cocoa [44, 45]. Decreasing blood pressure and cardiovascular mortality has also been demonstrated with cocoa consumption in a substudy of the Zutphen population [46]. Another study in smokers also demonstrated improved flow-mediated vasodilation after a cocoa rich diet [47]. An association of decreased cerebral perfusion with dementia has been recently highlighted [48] and the prospect of increasing cerebral perfusion with cocoa flavanols is extremely promising. A recent pilot study evaluated the relationship between CBF and a single acute dose (450 mg) of flavanol-rich cocoa and found this treatment to increase local CBF to grey matter by up to 60% by 2–3 h postconsumption [48]. Researchers from the University Hospital of Cologne pooled data from five studies regarding the effects of cocoa on blood pressure involving 173 participants and found that the consumption of cocoa had significant positive effects on reducing blood pressure [49].

Similar to cocoa containing drinks, another flavanol-rich food, dark chocolate (100 g containing 500 mg polyphenols) has been shown to be able to lower blood pressure and improve insulin sensitivity in healthy people [50]. In a randomised, sham procedure-controlled, crossover study, 100 g of dark chocolate (75% cocoa) eaten on two separate days [51] resulted in an increase in the diameter of the resting brachial artery, increased arterial blood flow and a significantly increased heart rate. This effect might be due to the presence of significant amounts of caffeine which can affect blood flow, heart rate and cognition independently of the polyphenol content.

There is no consistent correlations with the decrease in blood pressure and oxidative damage in humans. Recently Fraga [52] reviewed the mechanistic explanation to this paradox and highlighted the importance of membrane- or protein-flavan-3-ol interactions and related pathways.

6 Tea

After water, tea is the most widely consumed beverage worldwide. Tea contains flavones that can protect against CVD and may reduce the risk and lessen the outcome following CVA. A population-based study of 4 807 subjects over 5–6 years found an inverse association between tea consumption and the rate of incidental myocardial infarction [53]. An inverse relationship between tea intake and blood pressure [54, 55] and the prevalence of hypertension [55, 56] has also been reported. Further, a meta-analysis of studies on tea consumption and CVD risk, found that most studies demonstrate a decrease in the rate of myocardial infarction with increasing tea consumption, although the populations studied were very heterogeneous with strong geographical differences [57]. More robust inverse relationships were reported for continental Europe than from other areas; combining myocardial infarction and CHD as one outcome measure, three European studies found that the consumption of three additional cups of tea *per* day reduced risk by up to 66%. Green tea consumption in Japan has also been shown to be protective against coronary artery disease (CAD); a study of 512 Japanese patients found an inverse relationship between green tea consumption and CAD in men, but not in women [58]. A relatively recent human trial demonstrated an inverse relationship for both tea intake (525 mL) and 4-*O*-methylgallic acid (a tea derived polyphenol) excretion with blood pressure in older women [55]. Another recent meta-analysis demonstrated that tea polyphenols can positively influence blood pressure related disorders, although cocoa phenols were shown to have a greater efficacy [49]. These findings are consistent with the suggestion that long-term and regular ingestion of tea may have a beneficial effect on human blood pressure. This hypothesis was recently strengthened by the finding that longer-term green tea intake could prevent spontaneous stroke in an animal study [59].

7 Tomatoes

Various types of carotenoids may also help to prevent CVD. One human study of 28 subjects with an acute ischemic stroke found that the majority of plasma carotenoids are lowered immediately after such an attack, possibly as a result of oxidative stress [60]. A great deal of research has been conducted on the effects of lycopene and CHD [61, 62]. Further studies have shown that in addition to lycopene, polyphenols and other components of tomato products may be important in conferring protective vascular effects through its antioxidant action. It is interesting to note that processing of fresh tomatoes into an extract or paste can reduce the lycopene content by 9–28%, although the antioxidative potential appears to remain unchanged, or even to be increased as a result of processing [63]. Lycopene may also work in combination with other heart-healthy nutrients such as vitamin E, garlic or rosmarinic acid to inhibit LDL oxidation [64].

8 Soy

The cardiovascular benefits of naturally occurring diphenolic plant compounds, such as the isoflavones genistein, daidzein and its metabolite equol, have recently been examined in populations consuming high amounts of soy-based Asian foods. A clinical trial evaluating the benefits of soy isoflavones reported increased brachial artery FMD [65]. Clarkson [66] reported that plasma concentrations of genistein and daidzein range between 50 and 800 ng/mL in adults consuming soy-rich foods, similar to levels found in the Japanese population.

The cardiovascular benefits of soy products have been recognised by the US Food and Drug Administration (FDA) who allows a cardiovascular health claim to be linked to an intake of 25 g of soy protein *per* day. Soy protein in particular, has been shown to reduce levels of both cholesterol and blood lipids [67]. Extensive clinical and experimental evidence links hypertension and atherosclerotic vascular disease with the accumulation of oxidised LDL and the enhanced generation of reactive oxygen species within the vascular walls [68]. Cerebral vascular effects of soy metabolites such as equol are unknown. Recently, Jackman *et al.* [69, 70] have compared the vasorelaxant and antioxidant effects of equol and daidzein in the carotid and basilar arteries of normal and hypertensive rats. They concluded that in the normal condition, both equol and daidzein possess substantial vasodilatory and weak antioxidant activity in cerebral arteries, whereas in hypertension the vasorelaxant response to equol, but not daidzein, is preserved.

A progressive increase in oxidative stress and a generalised decline in antioxidant defences are common signs of ageing and age-related diseases, including vascular disorders. Although there is controversy concerning the benefits of soy isoflavones [71], it is clear that isoflavones can modulate vascular reactivity and have important anti-inflammatory roles *via* the activation of estrogen receptors and/or intracellular kinase signalling cascades [72]. The gender-specific bioavailability data of isoflavones and its metabolites is very important to formulate food composition/matrix characteristic of soy-based functional foods. A recent randomised crossover human trial has examined the effect of age, gender, and influence of the food matrix on the bioavailability of different soy foods [73], but more convincing human studies are required to make a definite health claims. Several human studies during last 10 years are not in agreement with the beneficial effect of soy isoflavones [74], rather the evidence favours soy protein. However, the American Heart Association Nutrition Committee has not ruled out the possibility that another component could be the active factor [74].

9 Pomegranate

There is abundant mythological and historical evidence regarding the health benefits of pomegranate fruit [75]. The fruit of the pomegranate (≈50% of total pomegranate weight) consists of 80% juice and 20% seeds. The fresh juice contains 85% water, 10% total sugars, 1.5% pectin, ascorbic acid and polyphenolic flavonoids [76]. The soluble polyphenol content varies between 0.2 and 1.0%, depending on the fruit variety and includes mainly anthocyanins, catechins, ellagic tannins and gallic and ellagic acids. Most research on its vascular effects has been carried out either using cell lines or animal models [77–80]. Some antiatherogenic properties of pomegranate juice (PJ) have recently been reported in human subjects. Results from a small randomised human trial suggested that PJ consumption (50 mL of PJ, containing 1.5 mM of total polyphenols) by patients with severe carotid artery stenosis induced a decrease in carotid intima-media thickness (IMT) and systolic blood pressure [80].

10 Do dietary polyphenols act as antioxidants *in vivo*?

Despite the strong indicative health benefits of plant polyphenols *in vitro* and in animal studies, their efficacy in humans is limited by several factors. First, the bioabsorption of polyphenols in the human gut, blood and brain is very low. The maximum plasma level of flavonoids (a class of polyphenol) in humans is usually reached between 1 and 3 h after the consumption of flavonoid-rich foods. These reach levels of between 0.06 and 7.6 μM for flavonols, flavanols and flavanones, and less than 0.15 μM for anthocyanidins [81]. In addition, the half-lives of flavonoids in human plasma vary depending on the food source but are generally in the range of few hours [81]. As well as being poorly absorbed, polyphenols are extensively metabolised in the intestine, liver and possibly brain tissue. This extensive metabolism of consumed polyphenols makes the use of *in vitro* assays measuring their effects prone to producing false positive results by illustrating effects of polyphenols which are not actually observed at bioactive levels in their native form in human plasma (*e.g*. quercetin, chlorogenic acid). Conversely, truly effective bioactivities from polyphenol metabolites may be missed because the native form was used *in vitro* and not the metabolites. Considering the relatively poor absorption rate and chemical modifications/conjugations occurring in the gut, blood and brain, one may have to consume many litres or kilos of these foods in order to achieve the necessary plasma molar ratios to neutralise free radicals and achieve the purported beneficial health effects. A new hypothesis is now being proposed, taking into consideration that the modest levels of polyphenols in the blood or brain are unlikely to be high enough to neutralise free radicals chemically, but rather, that these may stimulate complex intracellular signalling pathways leading to vascular protection aside from their direct chemical antioxidative properties. Thus the direct chemical free radical scavenging activity of dietary flavonoids in humans appears of limited importance [82, 83]. Recently, Schewe *et al*. [83] hypothesised that 'dietary flavonoids may act as antioxidants *in vivo* in a more broad sense by interfering with prooxidant processes or by inhibition of prooxidant enzymes such as NADPH oxidases, lipoxygenases and myeloperoxidase-mediated modifications of LDL'.

11 Conclusions

Foods and beverages rich in polyphenols are being heralded as potential preventive agents for a range of pathological conditions, ranging from hypertension to CHD, cerebral stroke and dementia. Recent human data from multiple laboratories on cardio- and cerebro-protective roles of plant polyphenols is very promising, especially concerning cocoa and cocoa products. We have followed a best-evidence scorecard in analysing peer-reviewed articles [84–89] as per Food Standards Australia New Zealand (FSANZ) guidelines [90]. Good to moderate evidences were found to support its health claims for GSEs, cocoa and dark chocolate (Table 1). The key question is whether these polyphenols can be used as both preventative and acute therapeutic agents in the treatment of cerebrovascular diseases. In a preventive approach, they may reduce atherosclerotoic lesions from forming in the first place, reduce thrombosis and lower blood pressure and cholesterol levels. Acutely, polyphenols improve CBF, prevent platelet aggregation and inhibit oxidative stress. Due to the pleiotropic properties of the polyphenols and their potential synergies of action on vascular endothelium when various polyphenolic compounds are combined, they appear to be good candidates for both the prevention and treatment of cerebrovascular diseases. These findings raise the possibility that products rich in polyphenols can be developed to help maintain healthy brain and vascular function throughout life.

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12 References

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