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Revisión

Bioactive substances with preventive effect in cardiovascular diseases

Juana Mulero[1], José Abellán[2], Pilar Zafrilla[1], Diego Amores[1] and Pilar Hernández Sánchez[1]

[1]Department of Food Technology and Nutrition. Universidad Católica San Antonio de Murcia. [2]Cardiovascular Risk Chair. Universidad Católica San Antonio de Murcia. España.

Abstract

The effect of diet on cardiovascular disease prevention has been widely studied for many years. Numerous studies have confirmed that diets rich in fruits and vegetables (Mediterranean diet) are beneficial to the cardiovascular system and various bioactive food components have preventive effect on chronic diseases such as cardiovascular disease. In this paper we review the effect of bioactive substances included in the group of flavonoids (catechins and proanthocyanidins, anthocyanins and isoflavones), stilbenes such as resveratrol, bioactive peptides, plant sterols and polyunsaturated fatty acids omega-3 on the cardiovascular system.

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Key words: *Cardiovascular disease. Catechins. Anthocyanidins. Anthocyanins. Resveratrol. Omega-3.*

SUSTANCIAS BIOACTIVAS CON EFECTO PREVENTIVO EN LA ENFERMEDAD CARDIOVASCULAR

Resumen

El efecto de la dieta sobre la prevención de las enfermedades cardiovasculares ha sido ampliamente estudiado durante muchos años. Numerosos estudios han corroborado que las dietas ricas en frutas y hortalizas (dieta mediterránea) resultan cardiosaludables y que diversas sustancias bioactivas que componen los alimentos tienen un efecto preventivo en diversas enfermedades crónicas como son las enfermedades cardiovasculares. En esta revisión vamos a tratar ciertas sustancias bioactivas, como son algunas incluidas en el grupo de los flavonoides (catequinas y proantocianidinas, antocianinas e isoflavonas), estilbenos como el resveratrol, péptidos bioactivos, esteroles vegetales y ácidos grasos poliinsaturados omega-3.

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Palabras clave: *Enfermedad cardiovascular. Catequinas. Antocianidinas. Antocianinas. Resveratrol. Omega-3.*

Introduction

The prevalence of cardiovascular diseases is increasing throughout the world; not only in developed countries but also, and more significantly, in developing countries.

In order to prevent its emergence and its consequences, it is necessary to decisively address the risk factors related to these diseases, such as cigarette smoking, dyslipidemia, obesity, diabetes, hypertension and, more recently, hyperhomocysteinemia.

Experts recommended to follow a healthy, varied and balanced diet as the best way to prevent certain diseases, ensuring good health and quality of life, and related eating disorders with the emergence of a large number of chronic illnesses and imbalances. These recommendations are based on associations which have shown between the consumption of plan foods, primarily fruits, vegetables, legumes and whole grains and their preventive effects on cancer and cardiovascular disease[1]. In fact, epidemiological studies have shown an inverse association between the prevalence of these diseases and the consumption of fruits and vegetables[2].

As a result of this situation, arise 'functional' foods: foods that in addition to its nutritional properties can provide health benefits to those who consume them, and which are increasingly more supported by the scientific evidence.

Bioactive substances which we will treat in this review are: flavonoids (catechins and procyanidins, anthocyanins and isoflavones). Stilbenes, resveratrol, bioactive peptides, plant sterols and polyunsaturated fatty acids omega-3.

Correspondence: Diego Amores de Gea.
UCAM Universidad Católica San Antonio de Murcia.
Campus de los Jerónimos, N.º 135.
Guadalupe 30107, Murcia.
E-mail: damores@ucam.edu

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Catechins and proanthocyanidins

Catechins and proanthocyanidins are included within the group of flavonoids and present structures derived from the flavan-3-ol skeleton. Monomers (catechins) are founded in nature or condensed among themselves to form oligomers or polymers (proanthocyanidins).

The distribution of the flavanols in the plant kingdom is very broad. They are mainly present in fruits, tea, cocoa and grains and less on vegetables. Their dietary intake is limited since it is in little edible organs and astringency characteristics associated. Still in Spain, it has been estimated a daily intake between 18 and 31 mg per person. It must be taken into account that the content varies depending on the variety and the richest tissues in flavan-3-OLS are skin and seeds[3].

There are several epidemiological studies that point to the existence of a negative relationship between consumption of rich flavan-3-ols foods (especially wine, tea, cacao and fruit) and the cardiovascular disease risk, even if the conclusions are not always coincident, and sometimes are even controversial[4].

The beneficial effects on health of the catechins and proanthocyanidins present in fruits and vegetables can be related with its antioxidant properties (inhibition of LDL oxidation), inhibition of platelet aggregation, modulation of endothelial function and antihypertensive properties[5].

Studies on experimental animals and isolated tissues have been demonstrated that proanthocyanidins has a vasorelaxant effect and this is dependent on the NO-cGMP route in combination with a hyperpolarization due to the activation of channels K Ca^{2+}- dependent[6]. Also, cocoa and wine proanthocyanidins inhibits platelet activation induced by epinephrine in *in vitro* models, as well as the inflammatory platelet dependent response, suggesting, therefore, its potential applications as antithrombotic and anti-inflammatory[7].

The most studied aspect in relation to this type of compounds is perhaps, the effect of catechins and proanthocyanidins on different markers of inflammation. It has been observed that the consumption of grape seed decreased levels of proinflammatory molecules as C-reactive protein (CRP), interleukin-6 and tumor necrosis factor alpha (TNF-alpha) and increased production of inflammatory cytokines such as adiponectin[8] in rats that showed a slight inflammatory condition. The epigallocatechin gallate, characteristic of the composition in green tea reduces vascular inflammation by of increasing nitric oxide (NO) synthesis, thereby blocking endothelial exocytosis[9].

Several studies in experimental animals and in vitro models with different cell models have shown the hypotriglyceridemic effects of the catechins and proanthocyanidins grape seed (cholesterol-1-Alpha-hydroxylase) CYP7A1 and SHP nuclear receptor-mediated, involved both in lipid metabolism[10]. Intervention studies in healthy or mild hypercholesterolemic volunteers it has proven the hypolipidaemiant effect of procyanidins from cocoa, with decreased of total cholesterol and oxidized LDL levels and increased HDL[11].

In a study of hypercholesterolemic subjects, supplementation with grape seed extract rich in proanthocyanidins significantly reduced oxidized LDL[12] level significantly, possibly as a result of the influence on the production of endothelial NO[13]. The mechanism of protection against the peroxynitrite attack in vascular cells takes place through deposition of procyanidins in coronary endothelial cells surface and it is favored by the endothelial relaxation mediated by the enzyme that synthesizes nitric oxide (NO-synthase)[14].

The green tea flavan-3-ols are antithrombotic because of the antiplatelet effects, rather than its anticoagulant effects. Antiplatelet activity is due to inhibition of intracellular biochemical pathway prior to exposure to GPIIb / IIIa. The green tea flavanols also inhibit the flow of the intracellular calcium ion induced by treatment with thrombin[15].

Anthocyanins

Anthocyanins are natural pigments responsible for the blue, purple, red and orange color of many fruits and vegetables. The main dietary sources of anthocyanins are red fruits, mainly berries (berries) and red grapes, cereals and purple corn, as well as vegetables (such as cabbage) sheet and red wine[16].

An intervention with relatively low doses of anthocyanins in patients with vascular diseases is associated with a significant reduction of ischaemia, blood pressure, lipids levels, and inflammatory condition[17,18]. Grape juice (10 ml/kg) inhibits significantly the platelet activity and experimental coronary thrombosis in vivo[19]. Clinical studies show little effect on pro-inflammatory markers in healthy individuals, however, a recently study shows a significant improvement in cardiovascular plasma biomarkers risk after supplementation with anthocyanins[20].

Thus, it described that anthocyanins are able to act on different cells involved in atherosclerosis development, one of the causes that trigger cardiovascular dysfunction. Anthocyanins have protective effects against the secretion of MCP-1, (involved in the development of the atheroma plaques) induced by transcription factors (TNF -α) in human endothelial cells[21]. The vascular endothelial growth factor (VEGF) is a very important pro-angiogenic and pro-atherosclerotic; even though it has been proven that these metabolites may prevent their expression of vascular tissues in smooth muscle[22]. In macrophages, mulberry anthocyanins inhibit the biosynthesis of nitric oxide in cell[23]. In addition, also exert an inhibitory role in platelet aggregation, without influence on the reactivity of platelets when faced with powerful antagonists such as collagen[24].

Isoflavones

They are a type of polyphenol group of flavonoids. Soy represents the biggest regular contribution of isoflavones. This food is highly consumed in Eastern countries thanks to marketing through various formats as part of functional foods it becomes more present in the diet of Western countries[25].

Some studies point out the role of isoflavones in the decrease in serum cholesterol levels. The Framingham study in postmenopausal women, related the intake of phytoestrogens (isoflavones and Llignans) with a favorable lower cardiovascular risk[26] metabolic profile, though, studies with the administration of isoflavones in supplements form are controversial to demonstrate its effects on circulating cholesterol levels[27].

The effect of the non-hormonal properties of isoflavones on the reduction of cardiovascular disease risk is of great importance. Several clinical studies show that isoflavones reduce the susceptibility of lipids oxidation[28] and, moreover, it has been observed that they can have similar effects to digitalis in the relaxation of coronary arteries through a mechanism that involves antagonism calcium channel[29].

Therefore, this study proves the capacity of these flavonoids to restore endothelial function in patients with severe endothelial dysfunction[30,31]. In addition it also showed that such this polyphenol supplements reduced c-reactive protein levels (inflammatory marker). Many studies have proven the close relationship between inflammation and endothelial dysfunction, and the anti-inflammatory effects of isoflavones could be one of the mechanisms responsible for the restoration of endothelial function[31].

Ecuol, colonic metabolite of daidzein and genistein has been proposed as a metabolite with preventive activity on cardiovascular diseases. These properties are linked to a strong antioxidant potential thanks to its ability to increase the NON-bioavailable by regulating to the decline in the production of $O_2^{-\bullet}$ and, therefore, preventing modification to a atherogenic particle LDL[32].

Thus, the inhibitory effect of ecuol on NO production and iNOS gene expression describes a possible mechanism responsible for the antiatherosclerotic effect of ecuol and soya isoflavones, which may be a therapeutic agent in vascular disease, especially in cerebral circulation[33].

Resveratrol and cardiovascular health

Resveratrol (3,5,4'- trihydroxystilbene) is a phenolic compound, belonging to the family of the stilbenes; normally found in trans form but it can also be found in the cis form. Found in peanuts and derivatives, blueberries, dark chocolate, grapes, grape juice and wine[34]. All wine is the main source of resveratrol in the diet, especially red wine. Resveratrol is a cardioprotective compound that acts at different levels, and hence, their effectiveness.

Antiplatelet *platelet*

There are several studies showing the antiplatelet effect of resveratrol both in vivo and in vitro, although the mechanisms by which resveratrol exerts this protective effect are complex and are not yet well enlightened[35,36]. A possible proposed mechanism is the preferential inhibition of COX-1 on COX-2[37], because of the balance of prostaglandins synthesized by two isoforms of the COX enzyme regulates vascular homeostasis. Under certain conditions, the inactivation of COX-1 by resveratrol is irreversible, and the platelets are unable to synthesize new proteins, which implies that a fleeting exposure to resveratrol may have effects *in vivo* (human platelet renewal time is 10 days)[38].

Vasodilator

The vasodilating ability of resveratrol has been attributed to its ability to stimulate channels Ca^{2+} and ^{+}K, and to the improvement of nitric oxide (NO) signaling in endothelium. This last activity is due to the inhibition of the activity of NADH/NADPH oxidase, allowing a reduction in the superoxide basal production, and consequently a decrease in the inactivation of the nitric oxide[39]. *In vivo*, resveratrol increases the expression of oxide nitric synthetase, endothelial (eNOS) as inducible (iNOS)[40].

Resveratrol might, therefore, increase the NO concentration through an increase of the expression of NO synthetase and decrease inactivation by radical free. This suggests that resveratrol could be a powerful protective *vivo* against ischemic damage during myocardial infarctions.

Modulator of the systemic inflammatory response

In this sense, it has been shown that resveratrol is able to inhibit the synthesis and release of proinflammatory cytokines, modify the synthesis of Eicosanoids, inhibit the activity of certain immune cells, or inhibit the action of transcription factors NFkB and protein-1 activator[41]. The suppressive effect on the activity of NFkB could explain the inhibition of the synthesis of pro-inflammatory cytokines such as TNF, IL-1 and IL-6 which has been observed in mononuclear cells after being incubated with trans-resveratrol[42].

There are studies which attribute also anti-inflammatory effects to resveratrol, including inhibition of the expression of the adhesion molecules[41].

Modulator of lipid metabolism

Experiments *in vivo* in rats, spontaneously hypertensive, prone to myocardial infarctions, show that resveratrol reduces markers of oxidative stress as glyca-

ted serum and 8- hydroxyguanosine in urine albumin[43]. These and other studies suggest that resveratrol can inhibit LDL oxidation between other macromolecules *in vivo*, if the mechanism of action is direct or indirect even is to be determined.

Bioactive peptides

Bioactive or functional peptides are defined as amino acid sequences inactive precursor protein inside, carrying on certain biological activities after his release by chemical or enzymatic hydrolysis. Usually, peptides are small (3 to 20 amino acids) released during the industrial food processing or during gastrointestinal digestion[44].

The main effects of the bioactive peptides on the cardiovascular system described are those relating to its antithrombotic and antihypertensive activity.

Antithrombotic *activity*

The major isolated peptides with antithrombotic activity are present in milk.

Certain peptides sequences of dairy peptide, called casoplatelins, are similar to the fibrinogen γ chain[45]. Found "in vitro" that these sequences are inhibitors of platelet aggregation and fibrinogen γ chain to the human specific platelet membrane receptor[46]. These peptides are capable of focusing on specific receptors located on the surface of platelets, thus preventing the thrombus formation. Other peptides have also antithrombotic activity, in a dose-dependent manner, inhibiting platelet aggregation induced by ADP due to homology with the fragment f (572-575) of the Fibrinogen α chain[47].

Antihypertensive activity

The most studied antihypertensive activity of bioactive peptides is the inhibition of the activity of ACE (angiotensin converting enzyme)[48-49].

Plant sterols

Sterols are compounds associated with variable proportion between 0,2 and a 2% lipids. Its basic structure is the cyclopentanophenanthrene condensed 4-cycle system. Sterols are compounds that can be free or esterified with fatty acids, mainly saturated fatty acids[50].

There is a large amount of experimental evidence that demonstrated that the plant sterols have an important hypocholesterolemic effect, reducing both the concentrations of total cholesterol and LDL cholesterol[51].

The most studied effect of plant sterols is their inhibition of intestinal absorption of cholesterol. Plant sterols, being more hydrophobic than cholesterol, can move the cholesterol of micelles of absorption, thus reducing it, in addition, the plant sterols could reduce the rate of cholesterol esterification in the enterocyte (affecting the activity of ACAT)[52] and, as a result, thus reducing the amount of cholesterol exported into the blood in the chylomicrons form.

The inhibition of cholesterol absorption produces an increase in the synthesis of LDL receptor[53] which increases the elimination of LDL and also the IDL from the circulation, and given that these are the precursors of LDL, thus descends in addition their production without being affected triacilglycerides and HDL cholesterol concentrations[54].

There is no conclusive data regarding the effect of plant sterols on the bile metabolism, although some studies suggest that they produce an increase in the excretion of bile acids, while that is not no effect in others[55].

Omega-3 Fatty Acids

Fatty acids are part of triglycerides, complex lipids and can esterify cholesterol. Polyunsaturated fatty acids (PUFAS) contain more than one double bond. There are two families: the n-6 PUFA (ω- 6) and the n-3 (ω- 3). The main n-6 fatty acid is linoleic acid (LA, 18:2 n-6), widely distributed in plants, mainly in oils of vegetable, such as corn, sunflower, and soybean seeds. It is a precursor of arachidonic acid (AA, 20:4 n-6) synthesized in mammals, and therefore present in foods of animal origin. Acid α- linolenic acid (LNA, 18:3 n-3) is the precursor of the long chain n-3 PUFA and predominates in dark green foliage, oils from seeds of flax, rapeseed, nuts, currant, and soy. The animals that live at the bottom of the sea are rich in eicosapentaenoic (EPA, 20:5 n-3) and docosahexaenoic (DHA, 22: 6 n-3), while algae and marine plankton are also sources of PUFA n-3[56].

Polyunsaturated fatty acids (PUFA) omega type 3, mainly present in fish oils, seem to play an important role as anti-inflammatory agents, antiarrhythmogenic and cardiovascular level protectors[57]. Linolenic acid (octadecatrienoic; C18:3n - 3) is the primary precursor of docosahexaenoic acid (DHA) and origin of certain prostaglandins, leukotrienes, and thromboxanes with anticoagulant, anti-inflammatory, vasodilator and antiplatelet activity (PGE3, PGI3, TXA4 and LTB5). The competition for the desaturase and liver elongasas (as well as placental and lactating mammary gland) to form DHA instead of arachidonic acid (AA), derived primarily from (octadecadienoic linoleic acid; C18:2n - 6; basically from seed oils) seems to be the fundamental physiological mechanism that would explain such actions[58]. Therefore it is suggested that substitution by AG omega-3 omega-6 AG would inhibit the synthesis of proinflammatory cytokines such as TNF-α, IL-1 and IL-2, and at the same time would

decrease the expression of adhesion molecules in vascular endothelium[59].

In addition n-3 linolenic acid has a cardioprotective effect, reducing significantly the c-reactive protein, in studies involving human[60].

Conclusions

In recent years growing scientific evidence demonstrating the relationship between food and health, particularly cardiovascular disease. Foods as well as providing nutrients, contain a series of non-nutritious substances involved in plant secondary metabolism: substances colorants (pigments), aromatic, growth regulators, natural protectors against pests and others that do not have a nutritional function classically defined, are not considered essential to human health, but which can have a significant impact on the course of some diseases, such as phytochemicals or bioactive substances.

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