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Jabuticaba as a Source of Functional Pigments

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ABSTRACT
The relatively high concentration of anthocyanins in the diet is of prospective importance to human health. Anthocyanins contribute greatly to the antioxidant properties of certain colorful foods, such as grapes and cranberries. Many studies in recent years have focused on the study of these functional pigments from different perspectives. The present review highlights recent studies on the health-promoting properties of anthocyanins. It presents latent anthocyanin sources and demonstrates the potentiality of an under-utilized non-conventional source widespread in Brazil called jabuticaba (*Myrciaria cauliflora*).
KEY WORDS: Anthocyanins, functional pigments, health-promoting properties, jabuticaba.

INTRODUCTION

Oxidative reactions in the human body have been appointed as the cause of diseases initiation and progression. The damage generated by free radicals and reactive oxygen species has been linked to some neurodegenerative disorders and cancers, and oxidation of low-density lipoprotein is a major factor in the promotion of coronary heart disease (CHD) and atherosclerosis (1).

Epidemiological evidences suggest that a diet high in fruits and vegetables plays an important hole in reducing the incidence of many oxidative and inflammatory diseases (2, 3). The reason of this accomplishment can be the coloration of these foods, in general, a rich source of many phenolic antioxidants.

The wish of a healthier diet allied with the increasingly concern of consumers over the use of synthetic additives in food has pushed the food industry to search for new sources of natural pigments (4). Anthocyanins are a type of functional pigments, responsible for a wide range of colors present in vegetables, flowers, fruits, and derived products. It is known that anthocyanin pigments act as strong antioxidants and anti-inflammatory, with antimutagenic and cancer chemopreventative activities (5). These bioactive properties have been already demonstrated in "in vitro" and "in vivo" studies (6) and an increase of publications in this area can be observed in the recent years.

Grapes and berries are well known for their antioxidant properties due to the presence of anthocyanins, many studies were done to extract and evaluate these compounds. The challenge for obtaining this class of pigments in industrial scale can be achieved by researching under utilized tropical fruits. Jabuticaba (*Myrciaria caulifora*) is a Brazilian fruit that can be potentially used as anthocyanins source because of their high content. It is denoted by many different names such as jaboticaba, guaperu, hipavuru, sabará or ybapuru (7).

The present work aims to highlight recent studies on the health-promoting properties of anthocyanins. As well as, present new potential anthocyanin sources obtained from non-conventional plants, giving special attention to a widespread fruit in Brazil called jabuticaba (*Myrciaria cauliflora*).

ANTHOCYANINS CHEMISTRY

Phenolic compounds are part of the secondary metabolism of plants and are of great importance for their survivor in unfavourable environment. They protect the species against adverse factors such as drought, UV radiation, infections or physical damage and regulate their development (8). A class of phenolic compounds easily found in the Plant Kingdom is the anthocyanins. They are water-soluble pigments that confer the bright red, blue, and purple colors of fruits and vegetables such as berries, grapes, apples, purple cabbage, etc (9).

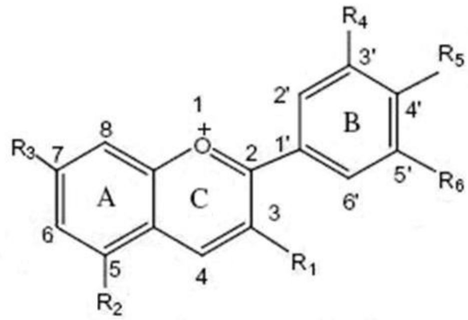
The basic structures of anthocyanins are the anthocyanidins (Figure 1). These structures, also known as aglycons, consist of an aromatic ring [A] bonded to an heterocyclic ring [C] that contains oxygen, which is also bonded by a carbon–carbon bond to a third aromatic ring [B] (10). When the anthocyanidins are found in their glycosylated form (bonded to a sugar moiety) via the C3 hydroxyl group in ring C they are known as anthocyanins.

In this way, a huge variety of anthocyanins can be observed spread in nature only varying in the basic anthocyanidin skeleton, the position and extent to which the glycosides are attached to the skeleton. The six most common anthocyanidin skeletons are cyanidin (Cy), delphinidin (Dp), pelargonidin (Pg), malvidin (Mv), petunidin (Pt), and peonidin (Pn) (Table 1) (when R1, R2, R3 and R5 are OH). Their distribution in fruits and vegetables is, respectively: 50%, 12%, 12%, 7%, 7% and 12% (11).

The most common anthocyanins in nature are the glycosylated derivatives of the three non-methylated anthocyanidins (Cy, Dp and Pg). They are found in 80% of pigmented leaves, 69% in fruits and 50% in flowers being the most common anthocyanin the Cy-3-glucoside (5).

ANTHOCYANINS HEALTH-PROMOTING PROPERTIES

Several studies show that a consumption of dietary phytochemicals, of which anthocyanins form a **considerable**



Name (when R1, R2, R3 and R5 are OH)	R4	R6
Cyanidin	OH	H
Delphinidin	OH	OH
Pelargonidin	H	H
Malvidin	OCH3	OCH
Petunidin	OCH3	OH
Peonidin	OCH3	H

Figure 1: Structural identification of anthocyanidins (aglycons)

Table 1: Sources of anthocyanins

Source	Anthocyanins (mg/100g of fresh weight)	References
Baguaçu	596,4-577,7	(33)
Bilberry	214.7-698	(34-36)
Blackcurrant	128-476	(37-39)
Black Bean	24.1-44.5	(40)
Black Olives	42-228	(36)
Black Rice	10-493	(41)
Blackberry	82.5-325.9	(42-44)
Blueberry	25-495	(34, 35, 45-48)
Cherry	2-450	(49)
Chokeberry	311.02-1480	(8, 34, 39, 50, 51)
Cranberry	19.8-140	(39, 51, 52)
Crowberry	360	(51)
Eggplant	8-85	(39, 51)
Jambolão	108,8-386	(33, 53)
Jabuticaba	310-315	(53)
Pomegranate (juice)	600-765	(54)
Port Wine	14-110	(38)
Purple Corn	1642	(55)
Raspberry	19-687	(39, 42, 44, 56, 57)
Red Cabbage	24,2-322	(39)
Red Grape	30-750	(29, 58, 59)
Red Onion	23.3-48.5	(39, 60)
Red Radish	100-154	(39, 61)
Rhubarb	4-200	(38, 51)
Strawberry	19-55	(62, 63)

Table 2: Articles that have mentioned and/or shown the potential of Jabuticaba as a source of functional pigments published in journals indexed in the Web of Science and Scopus database

Title	Year	Reference
Carbohydrates, Organic-Acids and Anthocyanins of *Myrciaria-Jaboticaba-Berg*	1972	(67)
Growth Relations and Pigment Changes in Developing Fruit of *Myrciaria Jaboticaba*	1996	(70)
Anthocyanin Antioxidants from Edible Fruits	2004	(1)
Application of Tristimulus Colorimetry to Optimize the Extraction of Anthocyanins from Jaboticaba (*Myricia Jaboticaba Berg.*)	2005	(4)
Blue Sensitizers for Solar Cells: Natural Dyes from Calafate and Jaboticaba	2006	(71)
Bioactive Depsides and Anthocyanins From Jaboticaba (*Myrciaria cauliflora*)	2006	(68)
Quantitative Analysis af Antiradical Phenolic Constituents from Fourteen Edible Myrtaceae Fruits	2008	(2)

part, may promote several health benefits. Due to their biological activity, in particular their antioxidant and anti-inflammatory activities, a reduction in the risk of cardiovascular disease, diabetes, cancer, an increase of the cognitive performance, and others can be achieved (12-15).

Prevention of Cardiovascular Diseases

Prevention of cardiovascular diseases is possibly the most studied effect of anthocyanins in the organism and the one for which a great quantity of epidemiological evidence exists. This class of phenolic compounds is capable of acting on different cells involved in the development of atherosclerosis, one of the leading causes to cardiovascular dysfunction (16).

Anti-diabetes Effects

According to some studies anthocyanins may also prevent type 2 diabetes and obesity. They affect the intestinal glucose absorption by retarding the release of glucose during digestion, insulin level/secretion/action and lipid metabolism "in vitro" and "in vivo". These phytochemicals were found to be potent inhibitors of starch digestion, and effective inhibitors of the a-glucosidase/maltase activity (17).

Anticancer Activity

Although it is not certain that anthocyanins intake reduces cancer risk in humans, the antioxidative capacity of these functional pigments is well known. Studies suggest that anthocyanins intake may reduce oxidative damage. A study performed in Germany showed that individuals who consumed an anthocyanins/polyphenolics-rich fruit juice had reduced oxidative DNA damage and a significant increase in reduced glutathione when compared to controls (18).

Anti-inflammatory Effects

Beneficial immune responses have been shown in human endothelial cells upon exposure to anthocyanins. The property of reducing the oxidative damage is the manly reason of these results, as inflammation processes are usually accompanied by excessive production of reactive oxygen and nitrogen metabolites. The anti-inflammatory effects could be confirmed by analyzing the compound metabolites at doses and comparing to those found in plasma after fruits anthocyanins-rich administration (19).

Protective Effect Against Hepatic Damage

Anthocyanins have shown to be effective in liver protection from hepatotoxicity induced by tert-butyl hydroperoxide (t-BHP) in studies with rats. These pigments decreased the serum levels of alanine and aspartate aminotransferase and reduced oxidative liver damage (20). In rats in which hepatic injury was induced by the administration of D-galactosamine the anthocyanins also demonstrated this protective effect (21).

Beneficial Effects in Cognitive Performance

Several studies performed in animals have shown that anthocyanins can increase the cognitive performance, and also protect the brain function by reducing oxidative ischemic damage and enhancing memory (13, 15, 22).

Protective Effect on Gastric Damage

The protective effect of anthocyanins on gastric damage is closely related with the capacity of this group of flavonoids to prevent or diminish the inflammatory process. It is known that inflammation implicates, at least initially, in processes of gastric injury. Studies have shown that cyanidin protects gastric mucosa from the damage caused by aspirin (23).

Cell Regeneration Properties

Mucopolysaccharides are important to maintain the integrity of both perivascular tissue and the basal membrane. Anthocyanins were reported to induce active phagocytosis of pigment material and intense cell regeneration in "in vitro" studies using endothelial cells from human umbilical cord (24). A growth promoting activity on fibroblasts and on smooth muscle cells was also reported in the same study. The regeneration of the cellular component of the vessel wall and of the perivascular tissues may be aid by anthocyanins intake, due to their stimulating effect on mucopolysaccharides.

Beneficial Ocular Effects

Anthocyanins have demonstrated a beneficial impact on the circulatory system improving the microcirculation of the blood and consequently improving vision at dusk and at night. Owing to those properties, anthocyanins have been applied in the production of ophthalmic preparations for research purposes (25).

Protective Effect Against Collagen Degradation

Elastase is an important proteolytic enzyme involved in the degradation of collagen and other components of the extravascular matrix in certain pathological conditions such as atherosclerosis, pulmonary emphysema, and rheumatoid arthritis. "In vitro" assays have demonstrated the ability of anthocyanins to inhibit these enzymes acting as a protection against collagen degradation. It is believed that anthocyanins interact with collagen metabolism by cross-linking collagen fibers and making them more resistant to collagenase action (26).

SOURCES OF ANTHOCYANINS

Pigments of plant materials have called the attention of scientists and the food industry in the last decades as source of extracts with biological properties. The coloration blue, red and purple found in many fruits, vegetables and leaves are of great interest since they are potential sources of anthocyanins extracts. This interest has increased lately, since, these pigments can be used as an alternative to artificial food colorants and also, because they are bioactive compounds.

Anthocyanins functions in plants are similar to the general functions of all flavonoids: antioxidant functions, photoprotectors, defence mechanisms, and other ecological functions (symbiotic phenomena). Since they give colour to different parts of plants, they also play an interesting role in the reproductive mechanisms: found in flowers, they serve to attract pollinators and in seeds and fruits to attract seed disseminators (27).

Anthocyanins pigments are usually extracted from plant materials with an organic solvent. The most common is ethanol and methanol containing a small amount of acid with the objective of obtaining the flavylium cation form, which is red and stable in a highly acid medium. However, acid may cause partial hydrolysis of the acyl moieties in acylated anthocyanins (28).

Recently, new techniques have been introduced for anthocyanins extraction from different sources, such as pressurized liquid extraction (29), sub and supercritical fluid extraction (30), ultrasound assisted extraction, high hydrostatic pressure or pulsed electric fields (31) and others.

The main sources of anthocyanins are red fruits, mostly berries and red grapes, cereals, mainly the purple maize, and vegetables (28, 32). Other potential sources of this nutraceutical (Table 1) should be analyzed for commercial proposes, since some plants can be found in great quantities and the extraction of the bioactive compound is facilitated as it is located in the external tissues of the plants.

Table 1 presents the concentration of anthocyanins from various sources using different extraction methods employing several solvents, and quantified by different anthocyanins quantification methods. The wide range of anthocyanins concentration obtained from the same source can be associated to the different extraction methods and also to different external and internal factors of the plant growth, such as genetic and agronomic factors, intensity and type of light, temperature and processing and storage of these agricultural matters (17).

Environmental conditions are known to induce the accumulation of anthocyanin pigments in the major groups of higher plants, light and temperature factors being the most studied ones. In the fruit of many crops such as grape, peach, strawberry, eggplants and lychee, anthocyanin synthesis is enhanced by sunlight and by cold weather (64, 65).

There are many other sources of anthocyanins-rich plants around the world such as fruits (in general their skins), flowers, stems, leaves and roots known and unknown until now. It is interesting to note the relevance of some under industrial utilized tropical fruits as potential source of anthocyanins. One of these is the Brazilian fruit called Jabuticaba (*Myrciaria cauliflora*).

JABUTICABA AS A POTENTIAL SOURCE IN BRAZIL

Jabuticabeira (*Myrciaria cauliflora* [Myrtaceae]) is a tree that grows mainly in Brazil, most frequently in the states of São Paulo, Minas Gerais, Rio de Janeiro, and Espírito Santo (66). This specie is often cultivated in home gardens, small-scale agricultural plots, or wild-harvested. They are primarily eaten fresh and can be found in local markets; they are also used to make jams, desserts, wines, liquors, and vinegars due to their short shelf life, usually 3 to 4 days after harvesting the fruits begin to ferment (7).

Jabuticaba is grape-like in appearance and texture, although its skin is thicker and tougher. This fruit has a dark purple to almost black skin color due to a high content of anthocyanins (310-315 mg/ 100 g of fresh weight) that covers a white gelatinous flesh inside (53, 67). It is 3 to 4 cm in diameter and carries from one to four large seeds. The fruits are born directly on the main trunks and branches of the plant, lending to a distinctive appearance to the fruiting tree (68).

Even with few studies reported in the literature and a yet not well known phytochemistry of this fruit, its sun-dried skins is traditionally used as a treatment for hemoptysis, asthma, diarrhea and chronic inflammation of the tonsils (69).

In a careful literature survey it was found only 7 articles that linked the jabuticaba fruit to its anthocyanins pigments published in journals indexed by the Web of Science and Scopus database (Table 2). Of these, only 4 mentioned and/or showed the potential of this Brazilian fruit as a source of functional pigments (Table 2).

The first one (reference number 67 of Table 2) was published in 1972 and its aim was to determine the concentration and type of anthocyanins in the jabuticaba jam process. The anthocyaninc extracts were purified and separated using thin layer chromatography. The chromatography and chemical properties (easy degradation) of these pigments supplied the first indication about the structure of the two isolated pigments (Peonidin and Peonidin-3-monoglucoside).

The other 2 papers (references 2 and 4 in Table 2) focused on the quantitative analysis of anthocyanins from jabuticaba.

In reference number 68 more attention was given to the identification of the anthocyanins present in jabuticaba and their antiradical activities. A new depside, jaboticabin, together with 17 known compounds were isolated from the jabuticaba skin in this study.

The last 3 papers (references 70, 1, and 71 in Table 2) focused on different aspects on the development of jabuticaba. The first reference (70) discusses the accentuated increase of anthocyanins concentrations at the end of the fruit growth cycle. The second one (1) evaluates the antioxidant activity of aqueous extract from jabuticaba. And the last paper (71) uses the jabuticaba skin extracts as semiconductor sensitizer for solar energy production. In this paper the results showed a successful conversion of visible light into electricity by using anthocyanins dye as wide band-gap semiconductor sensitizer in dye-sensitized solar cells.

CONCLUSION

This review has summarized some important papers that confirm that besides color, anthocyanins have properties that are beneficial to human health, with potential physiological effects such as anticancer, vasoprotective, anti-inflammatory, hepatoprotective, among others.

Tropical under utilized fruits as jabuticaba (*Myrciaria cauliflora*) in Brazil has demonstrated to be a good option of non-conventional sources of anthocyanins as natural food colorings due to their dark purple skins rich in anthocyanins (310-315 mg/ 100g of weight fresh). However, there is still a lack of information in the literature (just 4 papers published in journals indexed in the Web of Science and Scopus data) to promote this Brazilian fruit as a potential source of functional pigments.

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