Bioactivity and functionality of anthocyanins: A review

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Abstract

Anthocyanins are flavonoid compounds imparting pink, red, blue, and purple colors to fruits, flowers, leaves, and some underground organs. These pigments play an important role in pollination and fruit and seed dispersal and they are related to the protection of plant tissues exposed to biotic and/or abiotic stress. Their consumption has been linked to diverse health benefits such as anti-inflammatory and anti-carcinogenic properties, diminution of cardiovascular disease incidence, obesity control, and diabetes mitigation effects. Also, *in vitro* studies have revealed certain potential cancer chemopreventive activities of anthocyanins. Reactive oxygen species scavenging as well as metal chelating and direct binding to proteins have been associated with anthocyanin health effects. Recent studies have indicated that these compounds can modulate decisive signaling pathways and gene regulation. Their action contributes to vascular homeostasis and to angiogenesis, inflammation, and platelet aggregation control. Likewise, anthocyanins have been associated to the prevention of peroxynitrite-mediated disorder in endothelial cells. The potential use of anthocyanins as colorants in food has been extensively analyzed since they are the main hydrosoluble plant pigments. However, their instability and the influence of concentration, pH, temperature, and the presence of other compounds (phenolic acids, metals) on color properties can limit anthocyanin use as food ingredients and additives. Encapsulation has been proposed as an alternative to reduce food-processing damage and to deliver these compounds to their target site in the human body. Likewise, chemical lipophilization of anthocyanins has been assayed to obtain modified compounds with higher lipophilicity, more suitable for lipid-rich matrices.

Introduction

Amongst the diversity of natural products from plant origin, flavonoids represent one of the largest groups of phenolic compounds. Anthocyanins (from the Greek: *anthos*=flower and *kyanos*=blue) correspond to flavonoid pigments imparting pink, red, blue, and purple colors to fruits, flowers, leaves, stems, and some underground organs. Due to their water-soluble nature, anthocyanins accumulate in the vacuoles of a wide range of cells in both vegetative and reproductive tissues.

In 1915, the German chemist Richard Willstätter was awarded the Nobel Prize in Chemistry for his research on plant pigments. Besides his exhaustive research on chlorophyll pigments, Willstätter also studied anthocyanins. He isolated the characteristic pigments and investigated their chemical nature from many flowers and some fruits: cornflower, roses, pelargonia, larkspur, hollyhock, bilberries, black grapes, and cranberries [1]. He elucidated the chemical structure of these compounds and, by the reduction of the flavonol quercetin, he obtained the

cyanidin, which occurs in roses and cornflowers [2]. Willstätter was also able to chemically synthesize pelargonidin and showed the dependence of the flower pigmentation upon the reaction of the plant sap.

From an ecological point of view, anthocyanins provide attractive colors and aesthetic appeal to plants, playing an important role in pollination, and fruit and seed dispersal [3]. Different pigment colors are perceived by animals depending on their photoreceptors. Thus, color predilection is different for diverse pollinators. Many insects have restricted color vision on the red region of the electromagnetic spectrum and red coloration seems to play a major role in both bee-deterrence and bird-attraction [4]. Some butterfly and bird species visit anthocyanin-bearing red flowers, suggesting that both groups of animals are attracted by red anthocyanins [5]. Conversely, blue anthocyanins appear to attract bees more than the red ones.

The most widely accepted function for foliar anthocyanins is related to the protection of plant tissues exposed to biotic and/or abiotic stress [6]. Cell vacuole anthocyanins protect chloroplasts from photoinhibition and photooxidation in high light intensity environments and prevent the degradation of photolabile defense compounds. By scavenging free radicals and reactive oxygen species, anthocyanins lessen photooxidative damage in leaves [7].

Anthocyanins have been implicated in tolerance to stress factors such as drought, UV-B radiation, and herbivore and pathogen attacks.

Li et al., [8] reported that two *Arabidopsis* glycosyltransferase genes (UGT79B2 and UGT79B3), could be strongly induced by several abiotic stresses, including cold, salt and drought stress. The authors identified these enzymes' activities in the addition of UDP-rhamnose to cyanidin and cyanidin 3-O-glucoside, conferring abiotic stress tolerance via modulating anthocyanin accumulation.

Anthocyanin buildup induced by salinity stress has been reported in many species, such as sugarcane, strawberry, and rice. Elevated concentration of anthocyanins in response to high salinity environments has been demonstrated to confer high salt stress tolerance in *Brassica napus* plants with overexpressed *Arabidopsis* dihydroflavonol-4-reductase (DFR); increased DFR transcript levels correlated with higher anthocyanin accumulation [9].

According to Majetic et al., [10] the so-called phenotypic plasticity (i. e. the ability of organisms to differentially express their phenotypic traits depending on environment conditions) has been widely analyzed for an assortment of combinations between plant species and environmental factors. Nicotra et al., [11] have pointed out that anthocyanins are produced in leaves in response to an excess of light and temperature, and to osmotic extremes, and assist as a reversible, adaptable mechanism for the guard of photosynthetic machinery.

Ecological roles of anthocyanins can also be illustrated, for example, with the results found by Wang et al., [12] who reported greater concentrations of anthocyanins in *Vaccinium uliginosum* berries from higher altitude locations. This trend may be related to the protective mechanisms developed by plants against the increased intensity of solar radiation in more elevated areas. The authors also found a significantly positive correlation between anthocyanin contents and antioxidant activities measured by the DPPH, ABTS$^+$, and FRAP methods.

Other specific anthocyanin roles in plants are also generally accepted, such as contributors to cell redox homeostasis, mediators of signaling cascades induced by reactive oxygen species, chelating agents, and delayers of leaf senescence, particularly in plants suffering nutrient shortage [6]. Specifically, accumulation of anthocyanins in response to inorganic phosphorus deficiency has been reported in different plant tissues [13], and the genes involved in anthocyanin biosynthetic and regulatory pathways are upregulated during inorganic phosphorus deficit [14].

Structure, chemical characteristics and properties

The flavylium cation (an oxonium ion, namely 2-phenylbenzopyrylium) constitutes the basic structure of *anthocyanidins* [3], which are derivatives of the 3,5,7,4'-tetrahydroxyflavylium

cation (C15) [15] (Figure 1). This arrangement represents the aglycon component of the whole structure, which is known as *anthocyanin* when glycosylated. The presence of conjugated double bonds leads to the absorbance of visible light with a peak in the 465–550 nm wavelength zone. The degree of hydroxylation and the type and/or number of substituents determines the wide range of anthocyanin-derived colors. Another maximum absorption band falls in the UV range, between 270 nm and 280 nm.
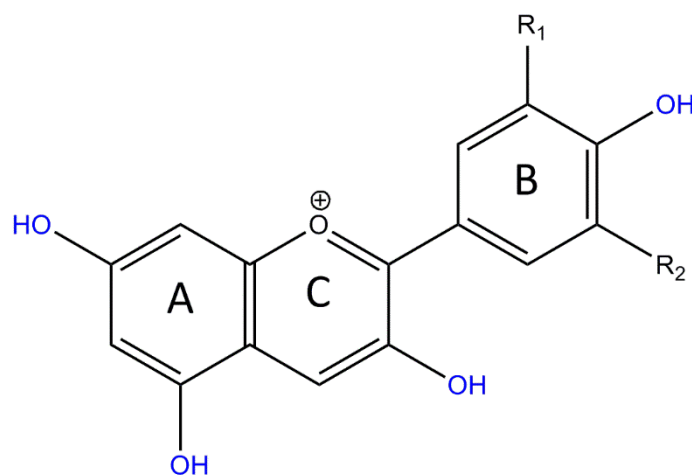

Figure 1. Chemical structure of anthocyanidins

Anthocyanins can be mono-, di-, or tri-glycosides, depending on the amount of sugar residues attached [3]. In the glycosyl moiety, most ubiquitous sugars are glucose, galactose, rhamnose, xylose, and arabinose. Rutinose has also been reported. Glycosylation usually takes place at the 3- and 5-hydroxyl positions for anthocyanidins [16]. In general, glycosidic substitution at position 3 yields a ratio between the absorbance value at 440 nm (E_{440}) and the one at the maximum visible absorption wavelength ($E_{vis-max}$) in the range of 20–35% [12].

Acylation may also occur: the sugar residues could be acylated by phenolic or aliphatic acids. Combination with cinnamic acids is common, which includes *p*-coumaric, caffeic, ferulic and sinapic acids as acylating moieties [17].

Although the different combinations allow more than 700 anthocyanin structures [18-19], there are 23 known anthocyanidins [20]. Amongst them, six different aglycon structures frequently found in nature are of most dietary significance: cyanidin, delphinidin, malvidin, pelargonidin, peonidin, and petunidin (Table 1).

Table 1. Most abundant anthocyanidins and their substituents groups -R_1 and -R_2.

Anthocyanidin	-R_1	-R_2	Color
Cyanidin	-OH	-H	Red
Delphinidin	-OH	-OH	Bluish purple
Malvidin	-OCH$_3$	-OCH$_3$	Reddish purple
Pelargonidin	-H	-H	Orange-red
Peonidin	-OCH$_3$	-H	Pink
Petunidin	-OCH$_3$	-OH	Purple

According to Burton-Freeman et al., [3] the color and stability of anthocyanins depend on their structure, the inter and intramolecular association with other compounds (e.g. ascorbic acid, metal ions, sugars, proteins), medium pH, temperature, light, oxygen, amongst others.

Regarding structural factors, anthocyanin color is influenced by the B-ring substitution pattern. The B-ring can be hydroxylated in different positions to yield different anthocyanin pigments

(Figure 1 and Table 1). These functional groups are introduced at an early stage in the metabolic pathway by different flavonoid hydroxylases. Pelargonidins, present for example in strawberries [21], provide orange, pink and red colors while cyanidins, present in relatively high proportions in dietary sources such as wild mulberries, blackberries, and raspberries [22] generate red and magenta tints. Delphinidins provide violet and blue colors (blue gentian *Gentiana triflora*, *Evolvulus pilosus*) [23-24]. Further color modifications arise from the introduction of methoxyl groups (catalyzed by *O*-methyl transferases) to form peonidin, petunidin, and malvidin (Table 1), which produce bright red, deep violet, and burgundy red, respectively.

On the other hand, betalains (a much smaller group of pigments with colors like those of anthocyanins) has received less attention. Betalains are tyrosine-derived compounds restricted to certain fungi and only one order of higher plants (the *Caryophyllales* order) [25]. Since betalains and anthocyanins are not naturally found together in any existing species, and because of their comparable optical and chemical properties, their role as functional homologues has been suggested. In this sense, betalains might be a substitute for anthocyanins in anthocyanin-free plants. Two structurally different types of betalains (Figure 2) have been described: a) the betacyanins (red in color, with maximum absorbance near 536 nm), which are the products of glycosylation and acylation of cyclo-dihydroxyphenylalanine; and b) the betaxanthins (colored yellow-orange, with maximum absorbance at approximately 470 nm), which are the result of the condensation of betalamic acid and assorted amino compounds. In the same way as anthocyanins, betalains are potent antioxidants and they can scavenge a variety of reactive species. These compounds have also a relatively high osmotic potential, which might give them the ability to serve as osmoregulators [25].
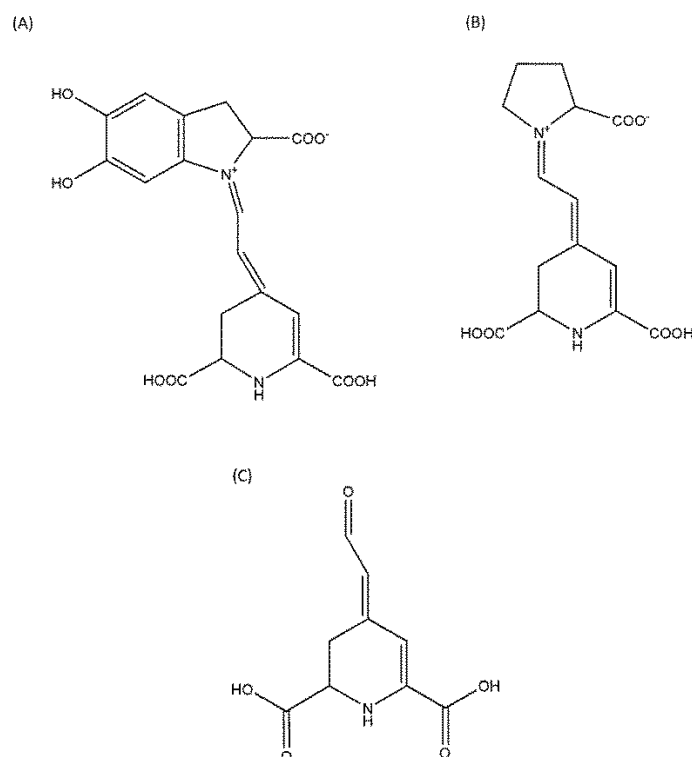

Figure 2. Chemical structures of: (A), a betacyanin (betanidin); (B), a betaxanthin (indicaxanthin); and (C), betalamic acid.

The generally accepted mechanisms for radical scavenging activity of phenolic compounds, which work as parallel reactions, are: 1) the donation of a hydrogen atom to the active radical giving a stable phenoxyl radical in one step (hydrogen-atom transfer mechanism); and 2) the

transfer of an electron and a proton in two consecutive steps, to give a radical cation and a phenoxyl radical, respectively (the single electron transfer mechanism) (Figure 3). In this sense, anthocyanins show high antioxidant activity, related with the capacity to scavenge free radicals by donating a hydrogen atom from a hydroxyl group as well as to support an unpaired electron [26]. Resonance phenomena and intramolecular hydrogen bonding can stabilize the phenoxyl radicals produced in this reaction. In this regard, Ali et al., [20] examined eight anthocyanidins, seven anthocyanins and two synthesized 4'-hydroxyflavyliums as hydrogen and electron donors. By means of a structure–activity relationship study, the authors reported that, in the absence of the 3-OH group, radicals of the 4, 5 or 7-OH groups can only be stabilized by resonance through pyrylium oxygen, whereas 3-OH group enhanced hydrogen atom donation due to the stabilization by anthocyanidin semiquinone-like resonance. Likewise, the 3-OH group enhanced electron donation.
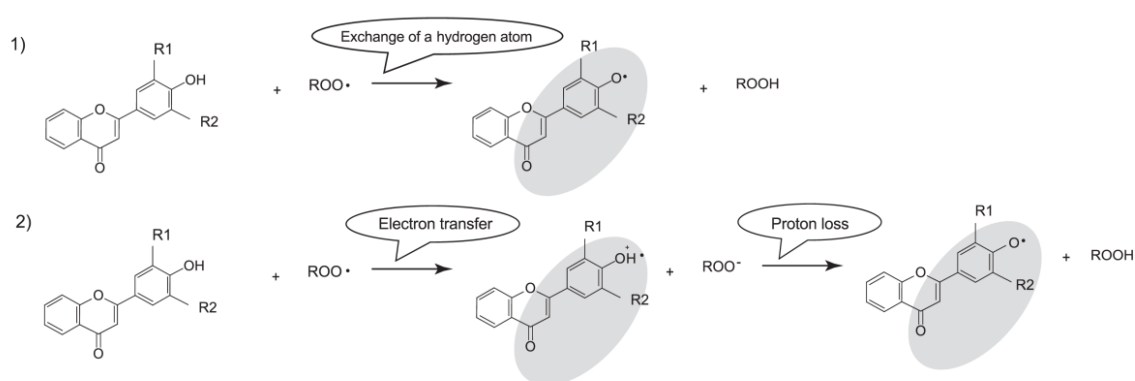

Figure 3. Accepted mechanisms for radical scavenging activity of phenolic compounds. 1) hydrogen-atom transfer mechanism; 2) single electron transfer mechanism. The shaded region on the phenolic molecules outlines the conjugated system stabilizing aroxyl radical or radical cation.

Bioactivity and health benefits

Consumption of anthocyanins has been linked to protective effects against many chronic diseases, due to strong antioxidant properties that lead to diverse health benefits, such as anti-inflammatory and anti-carcinogenic properties, diminution of cardiovascular disease incidence, obesity control, and diabetes mitigation effects.

Anthocyanins and their metabolites can prevent body fat accumulation and modulate inflammatory processes that appear to reduce the lipopolysaccharide (LPS)-induced nuclear factor-kappa B (NF-κB) translocation to the cell nucleus and mediate inflammatory responses [19, 27-30].

Recently, studies about the action mechanism of anthocyanins have indicated that these compounds can modulate decisive signaling pathways and gene regulation. Their action is localized at the vessel endothelium, contributing to vascular homeostasis and to angiogenesis, inflammation, and platelet aggregation control [31]. Likewise, anthocyanin capability to regulate enzymes involved in nitric oxide activity allows preventing peroxynitrite-mediated endothelial disorder in endothelial cells.

Zhang et al., [32] have studied the ability of anthocyanins from purple carrots and potatoes to ameliorate cellular inflammation in mono- and co-culture cell models. Potato anthocyanins seemed to be more bioaccessible in the *in vitro* gastrointestinal model used, and significantly

inhibited cellular inflammation in Caco-2 cells. Likewise, in an inflamed Caco-2 BBe1 plus human macrophage THP-1 co-culture cell model, transported anthocyanins could inhibit isoleukin (IL)-8 and tumor necrosis factor (TNF)-α secretion, and the expression of pro-inflammatory cytokines by blocking nuclear factor (NF)-κB.

The degree of efficiency of these properties was related to compound structural features. Results suggested that dietary anthocyanins derived from the selected purple underground vegetables, particularly purple potatoes, might contribute to restore mucosal homeostasis in inflamed gut.

Likewise, the anti-inflammatory and anti-proliferative activities of anthocyanins from cocoplum (with delphinidin, cyanidin, petunidin and peonidin derivatives identified as the major extract components) were assessed in CCD-18Co non-malignant colonic fibroblasts and HT-29 colorectal adenocarcinoma cells [33]. Cell proliferation was suppressed in HT-29 cells at 10.0 and 20.0 μg mL^{-1} of gallic acid equivalents, accompanied by decreased TNF-α, IL-1β, IL-6, and NF-κB1 expressions at the higher concentration. The authors suggested that cocoplum (*Chrysobalanus icaco*) anthocyanins have cancer-cytotoxic and anti-inflammatory actions in both inflamed colon and colon cancer cells.

In a recent study, capuli (*Prunus serotina* subsp. capuli) fruits proved to be a natural source of bioactive anthocyanins, since nitrites, the interleukin 1β messenger RNA, and the tumor necrosis factor α mRNA levels were significantly reduced when RAW 264.7 macrophages were incubated with different concentrations of capuli crude extract and then stimulated with lipopolysaccharide (LPS), to determine the markers related to oxidative damage and the proinflammatory cytokine production [34].

Although anthocyanins have been increasingly investigated for their neuroprotective and antineuroinflammatory effects, some research has been oriented to evaluate if phenolic acids (derived from anthocyanin metabolism) might be the bioactive constituents of anthocyanin-rich diets, considering that the overall bioavailability of many anthocyanins is relatively low. Winter et al., [35] examined the neuroprotective capacity of 4-hydroxybenzoic acid (HBA) and protocatechuic acid (PCA), in primary cultures of cerebellar granule neurons. Both HBA and PCA could mitigate oxidative stress induced by H_2O_2, a reactive oxygen species which is supposed to play a part in neuronal cell death and neurodegeneration. During nitrosative stress, only PCA acted as neuroprotective, while HBA did so under conditions of excitotoxicity. The authors also analyzed the anti-inflammatory activity of HBA and PCA in microglial cells stimulated with lipopolysaccharide, being PCA an effective anti-inflammatory agent by the reduction of nitric oxide production.

It is worth noting that Phase I Clinical trials recently demonstrated the ability of black raspberries to modulate inflammatory biomarkers of molecular efficacy that suggests a chemopreventive strategy against oral cancer [36]. In consequence, the authors established the ability of black raspberries to inhibit lesion formation in a rat model of 4-nitroquinoline 1-oxide (4NQO) induced oral carcinogenesis, and they examined the molecular efficacy using a clinically relevant transcriptional signature. The results allowed to conclude that dietary administration of black raspberries inhibited oral carcinogenesis in the rat 4NQO model [36].

Regarding the roles of anthocyanins on the prevention and treatment of cancer, Lin et al., [37] have reviewed the probable antitumor or anticancer effects of these compounds in the different stages of tumorigenesis and carcinogenesis. These actions are reported to be based on anthocyanin bioactivities, which include antioxidant properties, anti-inflammatory activity, anti-mutagenic effects, proliferation inhibition (by modulating signal transduction pathways, inducing cell cycle arrest, and stimulating apoptosis or autophagy of cancer cells), anti-metastasis action, reversion of drug resistance of cancer cells, and/or increasing their sensitivity to chemotherapy.

To assess if anthocyanins and their metabolites (isolated from plasma of healthy volunteers after ingestion of an anthocyanin-rich juice) were effective in modulating cancer cell migration *in vitro*, Kuntz et al., [38] analyzed the migration of pancreatic cancer cells PANC-1 and AsPC-1,

cellular or mitochondria-specific reactive oxygen species, and mRNA expression levels of matrix metalloproteinases (MMP-2 and MMP-9) and NF-κB mRNA. After application of isolated plasma anthocyanin metabolites to PANC-1, a reduction of cell migration was observed, with simultaneous lower levels of endogenously generated reactive oxygen species, and reduced NF-κB, MMP-2 and MMP-9 mRNA expression levels. Nevertheless, migration in AsPC-1 cells was not influenced by plasma anthocyanin metabolites. Thus, anthocyanins and their metabolites derived from an anthocyanin-rich juice might inhibit pancreatic cancer cell migration, depending on the cell phenotype.

Likewise, the effect of a bilberry anthocyanin-rich extract on colorectal tumor development and growth induced by azoxymethan/dextran sodium sulfate in female Balb/c mice was evaluated [39]. Added to a significantly lower mean tumor number, mice fed with 10% anthocyanins in the diet showed significantly longer colons as compared to controls, which represents an indication of reduced inflammation. In the same sense, diets containing freeze-dried black raspberries suppressed the development of N-nitrosomethylbenzylamine–induced tumors in rat esophagus [40]. Besides, the residue of black raspberries appeared to be as effective as the anthocyanin fraction in preventing esophageal cancer in rats.

Mazewski et al., [41] have evaluated the anti-proliferative effect of anthocyanin-rich purple and red corn on HCT-116 and HT-29 human colorectal cancer cells, finding that the assayed extracts increased apoptotic cells and impacted markers of apoptosis and angiogenesis. All corn extracts decreased the expression of vascular endothelial growth factor. Purple and red corn extracts could promote apoptosis and suppress angiogenesis and they might potentially inhibit human colon cancer cells.

Limsitthichaikoon et al., [42] carried out the complexation of anthocyanins from aqueous extracts of dried *Zea mays* cobs and *Clitoria ternatea* petals and evaluated the *in vitro* bioactivity against *Candida albicans* and their anti-inflammatory effect. Besides a better thermal tolerance, the complex was not cytotoxic (up to 2.5 mg/mL) to the tested human gingival epithelial cells and allowed to recover cellular proteins and nucleic acids of TNF-α induced inflamed HGEPp0.5 cells. Furthermore, minimum inhibitory and fungicidal concentrations of the anthocyanin complex against *C. albicans* were 80 and 160 µg/mL, respectively.

Likewise, Priprem et al., [43], have developed an anthocyanin complex for its use as a topical sunscreen. The main components of the product were the aqueous extracts of *Clitoria ternatea* and *Rosa damascena*. The authors showed that the formulation had no cytotoxic effect on human keratinocytes and fibroblasts in a concentration up to 1 mg/mL. The anthocyanin complex enhanced the stability of the pigments and diminished the *in vitro* permeation of total reducing contents through porcine skin, useful behavior for a better UV-A protection.

Regarding cardiovascular effects, Cassidy [44] has extensively reviewed the information obtained from observational studies concerning to the association between dietary anthocyanin intakes and cardiovascular disease risk. Some of them bring evidence about the increased usual anthocyanin intake being significantly associated with a reduction of 12-32% in the risk of coronary heart disease, in multivariate analyses. Likewise, prospective studies provided support for the observed decrease in coronary heart disease risk accompanying increased anthocyanin intake, by the improvement of arterial stiffness and blood pressure [44].

Reactive oxygen species scavenging ability as well as metal chelating and direct binding to proteins properties have also been associated with anthocyanin health effects [45].

In vitro analyses have revealed certain potential cancer chemopreventive activities of anthocyanins, including radical scavenging activity, stimulation of detoxifying enzymes, reduction of cell proliferation, inflammation, angiogenesis and invasiveness, and induction of apoptosis and differentiation [45].

On the other hand, Bhaswant et al., [46] have measured the main cardiovascular, liver and metabolic parameters in rats with diet-induced metabolic syndrome, following continual administration of anthocyanins, either from chokeberry (*Aronia melanocarpa*) or purple maize (*Zea mays*). The authors have found that rats fed with a high carbohydrate and fat diet, together with chokeberry juice or purple maize flour, showed reduced visceral adiposity index, total body fat mass and systolic blood pressure, enhanced glucose tolerance, liver and cardiovascular structure and function, and decreased plasma triglycerides and total cholesterol, compared to control animals. These findings suggest that anthocyanin supplementation from natural sources might be of assistance in attenuating obesity and metabolic syndrome.

Jennings et al., [47] have examined the relationship between usual intake of flavonoids, insulin resistance, and associated inflammatory biomarkers in women. To that effect, fasting serum glucose, insulin, high sensitivity C-reactive protein (hs-CRP), plasminogen activator inhibitor-1, and adiponectin concentrations were measured. The authors found that higher anthocyanin intake was associated with significantly lower peripheral insulin resistance due to a decrease in insulin levels, and lower hs-CRP concentrations. Anthocyanin-rich products were also related to lower insulin and inflammation levels.

Regardless of the biological activities that have been linked to anthocyanins, these compounds seem to be rapidly absorbed and eliminated [48]. Thus, the study of dietary anthocyanin bioavailability is a relevant issue. Amongst other authors, Stalmach [17] and Fernandes et al., [48] have extensively reviewed this topic.

Multiple factors affect the bioavailability of a compound, such as the amount ingested, presence of sugar moieties, composition and characteristics of the food matrix, co-ingestion of other compounds, rate of gastric emptying, intestinal mobility, dynamics of the fluids in the gastrointestinal tract, fluid secretion rate, as well as the transit time in the stomach, small intestine and colon [17, 49]. Metabolic reactions concerning anthocyanins comprise methylation, sulfation, conjugation with glucuronic acid, breakdown of glycoside linkages, and cleavage of the anthocyanin heterocycle.

According to Fernandes et al., [48] anthocyanins are readily detected in plasma in their parent forms after ingestion, probably because of their absorption through the gastric wall. Most polyphenols remain relatively stable at the stomach´s typical low pH and resist acid hydrolysis, which facilitates their transit to the small intestine without modifications. However, only anthocyanin glycosides are absorbed intact in both the stomach and small intestine. The absorption rate is limited by the type of sugar moiety attached [50].

Talavéra et al., [51] have found that rats subjected to gastric administration of the anthocyanins from bilberry extracts showed a rapid absorption of these compounds through their gastric lumen. The glycosides of delphinidin were most efficiently absorbed; while cyanidin, peonidin and petunidin-glycosides were absorbed with lower efficiency.

More recently, Atnip et al., [52] have studied the behavior of anthocyanins at pH 3.0–7.4 in an *in vitro* model of the gastric epithelium. The authors examined the effects of time, concentration, and pH on the carrying and uptake of anthocyanins, which were transported from the apical to the basolateral side of NCI-N87 cells in time and dose dependent ways. The results of this work support the hypothesis that anthocyanins are transported through gastric tissue by means of active transport mechanisms [52].

Likewise, using an *in vitro* model of mouse gastrointestinal tract, Matuschek et al., [53] demonstrated differences in cyanidin-3-O-glucoside absorption by different intestinal segments, with the highest absorption occurring in the jejunum, minor absorption observed in the duodenum and practically no absorption taking place in the ileum or colon.

On the other hand, Mueller et al., [54] compared the bioavailability of anthocyanins in healthy subjects versus ileostomists, which revealed significantly higher amounts of anthocyanins from bilberries and their degradants in the plasma/urine of subjects with an intact gut. The authors also pointed out that anthocyanins reached the circulation mainly as glucuronides. The

evidence of this work suggested that the colon is a significant place for the absorption of anthocyanins and their degradation products.

The ingested anthocyanins do not reach the gut microbiota in their original chemical forms. The respective metabolites excreted in the bile and/or from the enterohepatic circulation are finally available. In the colon, anthocyanins are metabolized by bacteria that release simpler compounds.

According to Stalmach, [17] once they reach the gastrointestinal tract, the chemical equilibrium of the ingested anthocyanins is modified, which may alter their sensitivity to degradation because of the different stability of the related species. However, the identification of some of the pH-related transformed compounds needs to be assessed by proper analytical techniques.

Jamar et al., [19] have pointed out that anthocyanins should be considered to have prebiotic activity, since these bioactive metabolites have been recognized as modulators of gut microbiota that contribute to obesity control.

Anthocyanin pigments are related to advantageous modification of the gut microbiota, since these compounds promote the colonization by bacteria (i.e. *Bifidobacterium spp.*, *Lactobacillus spp.*, *Akkermansia muciniphila*) that participate in provitamin and phenolic activation and absorption, gut barrier function improvement, mucus secretion stimulation, and intestinal immune response modulation [19]. In this sense, Pan et al., [55] reported that whole black raspberries (*Rubus occidentalis* Munger variety) and their residues have induced changes in male F344 rat gut bacteria, increasing the abundance of certain species recognized for their anti-inflammatory effects (i.e. *Akkermansia* and *Desulfovibrio*). Likewise, butyrate-producing bacteria such as *Anaerostipes* were also increased by whole black raspberries. Particularly, the black raspberry anthocyanin fraction was associated with an augmented abundance of *Anaerovorax* and *Dorea*, decreased population of *Bifidobacterium* and *Lactococcus* (weeks 3 and 6 of the assays), and a transiently increased abundance of *Asaccharobacter* and diminished population of *Prabacteroides* (week 3) [55].

Figure 4 summarizes the main bioactive properties and health benefits of anthocyanins reported from *in vitro* and *in vivo* assays, clinical trials and/or observational studies.
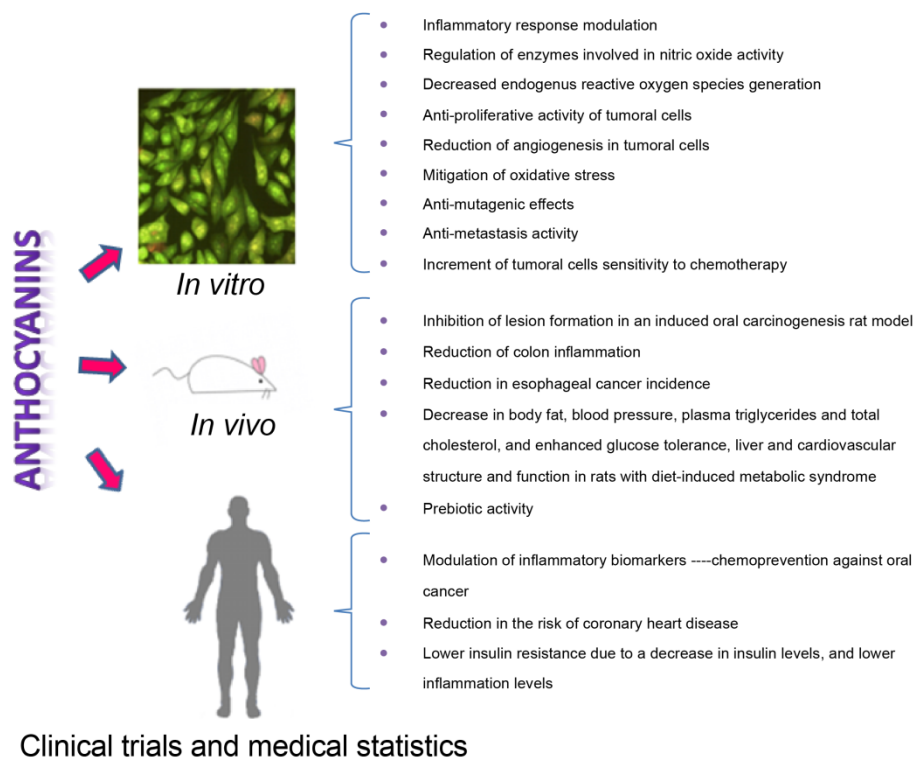
Figure 4. Summary of the main bioactive properties and health benefits reported for anthocyanins.

Anthocyanin sources

Anthocyanins can be found in many plant sources such as cereals, bulbs, tubers and roots, vegetables, pulses, and fruits. The relative richness of anthocyanins varies significantly depending on plant species and timing of harvest [56].

Table 2 shows common and potential plant sources of anthocyanins and their content range. Important sources of anthocyanins are fresh, red/blue fruits such as cherries, strawberries, plums, blackberries, raspberries, grapes, and red and black currants, where these pigments are mainly found in the skin, although they can also appear in the flesh. For example, red-fleshed sweet cherries (*Prunus avium*) are outstanding sources of anthocyanins [57].

Table 2. Some edible sources of anthocyanins

Plant source	Scientific name	Main identified compounds	Content range	Cultivars/ Commercial varieties	References
Black currants	*Ribes nigrum*	cyanidin 3-glucoside, delphinidin 3-glucoside, cyanidin 3-rutinoside, and delphinidin 3-rutinoside	Total anthocyanins, expressed as milligrams of cyanidin-3-glucoside equivalents per 100 g of dry extract: 207.5±8.89 - 372.9 ± 5.47	'Ben Lomond', 'Ben Sarek', 'Tsema', 'Titania', 'Čačanska Crna', 'Tisel', 'Tiben'	[58-61]

European elderberries, black or common elders	*Sambucus nigra*	cyanidin 3-sambubioside-5-glucoside, cyanidin 3,5-diglucoside, cyanidin 3-sambubioside, cyanidin 3-glucoside and cyanidin 3-rutinoside	603 – 1265 mg/100 g fresh weight	Cultivars Haschberg and Rubini; selections 13, 14 and 25	[62]
Blackberries	*Rubus spp*	Predominately cyanidin based in the non-acylated form	---	---	For anthocyanins reported in *Rubus* fruit, see [63]
Bilberries	*Vaccinium myrtillus*	Major anthocyanin moiety: delphinidin	Total monomeric anthocyanin range: 206.18 - 867.52 mg/100 g dry weight	Colored bilberry from six different Locations in Finland	[64]
Black chokeberry	*Aronia melanocarpa*	---	Total anthocyanin content, calculated as cyanidin-3-glucoside: 141±9 - 147±17 mg per 100 g of dry matter	---	[65]
Blueberries	*Vaccinium spp*	Galactoside and arabinoside derivatives of delphinidin, petunidin and malvidin aglycones (92% of total detected anthocyanins)	1435 mg of delphinidin-3-O-galactoside equivalents/100 g dry fruit (sample homogenized in a domestic mixer) - 2602 mg of delphinidin-3-O-galactoside equivalents /100 g dry fruit (Ultraturrax processed sample)		[66]
Açai berries	*Euterpe*	Main	Total		[67-68]

		anthocyanins: cyanidin-3-O-glucoside and cyanidin-3-O-rutinoside	anthocyanin content: 96.58 ± 0.11 mg/100 g of n lyophilized açaí berry pulp		
oleracea					
Juçara berries	*Euterpe edulis*	---	Total monomeric anthocyanins (mg cyanidin 3-glucoside equivalents 100 g^{-1} fresh matter):	---	For anthocyanins reported in *Euterpe* fruit and pulp, see [69]
Red raspberries	*Rubus idaeus*	Main Anthocyanins: cyanidin-3-O-sophoroside, cyanidin-3-O-(2"-O-glucosyl) rutinoside, cyanidin-3-O-glucoside and cyanidin-3-O-rutinoside	Total anthocyanins: 97.3 µmol/100 g	Cultivar Glen Magna	[70-71]
Purple-hulled Sunflower	*Helianthus annuus*	cyanidin-3-glucoside and cyanidin-3-xyloside	0.23-2.47 percent	Neagra du Cluj genotype	[72]
Red apples	*Malus × domestica* Borkh.	Cyanidin-3-O-hexoside, cyanidin-3-O-pentoside, B-type vitisin peonidin-3-O-hexoside, delphinidin-(epi)catechin adduct	-----	Red-fleshed apples from two regions of Iran: Bekran and Bastam	[73]
Red grapes	*Vitis vinifera* L.	Main anthocyanins: delphinidin 3-glucoside, petunidin 3-glucoside, peonidin 3-glucoside, malvidin 3-glucoside, malvidin 3-	30 to 750 mg/100 g of ripe berries	Varieties Cabernet Sauvignon, Merlot, Syrah and Tempranillo	[74]

		monoglucoside-acetate, malvidin 3-monoglucoside-p-coumarate			
Radish	*Raphanus sativus* L.	Main anthocyanins: pelargonidin-3-sophoroside-5-glucoside, mono- or di-acylated with cinnamic and malonic acids	12.2 - 53 mg per 100g root	Red-fleshed Winter cultivar	[75]
Eggplant	*Solanum melongena*	Mainly delphinidin derivatives, which accumulate in the peel	Total monomeric anthocyanins: 3.9 – 161 mg cyanidin-3-glucoside/100 g dry weight	Monarca and Perla negra	[76-77]
Red cabbage	*Brassica oleracea* L.	Main anthocyanins: cyanidin-3-diglucoside-5-glucoside non-acylated, and mono or di-acylated with sinapic, *p*-coumaric and ferulic acids	1111 to 1780 mg cyanidin-3-glucoside /100 g dry matter	Cultivars Cairo, Kosaro, Integro, Buscaro, Azurro, Primero, and Bandolero	[78]
Plums	*Prunus domestica* L.	Cyanidin 3-xyloside, cyanidin 3-glucoside, cyanidin 3-rutinoside, peonidin 3-rutinoside, peonidin 3-glucoside	36.6 – 7.4 mg/100 g FW	Cultivars 'Jojo', 'Valor', 'Čačanska rodna' and 'Čačanska najbolja'	[79]
Strawberries	*Fragaria× ananassa* Duch.	Main anthocyanins: cyanidin -3-glucoside, pelargonidin -3-glucoside, pelargonidin -3-rutinoside	200 – 600 mg kg^{-1}	Cultivars Eris, Oso Grande, Carisma, Tudnew and Camarosa	[80]
Sweet	*Prunus*	Cyanidin-3-	0.61 - 1.49	Lapins,	[57]

cherries	*avium*	glucoside, cyanidin-3-rutinoside, pelargonidin-3-rutinoside, peonidin-3-glucoside, peonidin-3-rutinoside	mg/g dry weight	Staccato, Sweetheart and Stella red-fleshed cherries	
Red wine	Wine from *V. vinifera* varieties	Main anthocyanins: malvidin-3-glucoside, malvidin-3-acetylglucoside, malvidin-3-*p*-coumaric glucoside	16.1 – 508 mg/L	Varietals Vranec, Merlot and Cabernet Sauvignon	[81]
Black olives	*Olea europaea L*	Main anthocyanins: cyanidin-3-rutinoside and cyanidin-3-glucoside	cyanidin 3-glucoside: 0.66 mmol/kg dry basis, and cyanidin 3-rutinoside: 1.27 mmol/kg db	Hojiblanca cultivar	[82-83]

Berry fruits are commonly consumed in fresh and processed forms, such as frozen or canned, but also as ingredients or derived products used in the elaboration of yogurts, beverages, jams and jellies [65]. Additionally, the intake of berry extracts as components in functional foods and dietary supplements is markedly increasing, in some cases combined with other fruits, vegetables and herbal extracts [65].

During fruit development, from fertilized ovary to full ripe fruit, a wide series of molecular and physiological changes take place, which are associated with color, cell wall softening, synthesis of specific volatiles, and modifications in the sugar/acid equilibrium. Fruit anthocyanin content can be affected by many factors, including genetic variation, developmental and environmental factors, and plant growth regulators [84].

When analyzing the concentration of anthocyanins in ten developmental stages of strawberry cultivar Selva, Ferreyra et al., [85] found that from the 50% red stage on, anthocyanin content increased exponentially and reached, at the purple-red stage, values 100 times higher than those for the white developmental degree.

Red coloration varies among fruit species and cultivars, and environmental conditions. For example, Asian pears, which mostly change their color at a late developmental stage, are greatly affected by light and temperature [86].

In non-climacteric fruits such as grape, strawberry and sweet cherry, it has been suggested that exogenous abscisic acid (ABA) up-regulates genes related to ripening, including those associated with cell wall changes and anthocyanin biosynthesis [87-89]. Jiang and Joyce [90] have pointed out that the phenylpropanoid pathway is activated by exogenous ABA and the anthocyanin content is increased in strawberries by the action of this phytohormone. As a result, red color development was accelerated in ABA treated fruits.

Likewise, other plant growth regulators, such as indole-3-acetic acid and methyl jasmonate are related to the inhibition or increase of flavonoids in red raspberries [84].

Red wine is also a good source of anthocyanins, with approximately 200–350 mg per liter. The early purple-red color of young wines derives from the anthocyanins extracted from grape skins, whereas this color changes during ageing to a more reddish-brown hue, mainly due to progressive structural changes of anthocyanins [91]. As the wine ages, anthocyanins can be transformed into more complex structures such as anthocyanin-pyruvic acid adducts and vinylpyranoanthocyanin-catechins [92].

According to Escribano-Bailón et al. [93], blue, red and purple varieties of some cereals are also potential sources to obtain anthocyanin-rich extracts, such as purple corn, which is a *Zea mays* pigmented variety mainly cultivated in Peru and Bolivia. Anthocyanins are also present in red and black colored-husk rice grains (*Oryza sativa*), which have been traditionally consumed in Asian regions. Likewise, the red color of sorghum (*Sorghum bicolor*) accessions has been associated to the levels of anthocyanins, in spite of its pericarp color being attributed to both anthocyanins and carotenoids [94].

According to Fernandes et al., [48] the estimation of the daily intake of polyphenols depends on several factors. The creation and application of a database with food polyphenol content has simplified this complex task [95-96]. In 2009, the first release of PhenolExplorer (Phenol-Explorer 1.0) has helped to assess polyphenol intake through an estimation of the polyphenol composition of a queried food [96]. The latest version (Phenol-Explorer 3.6) has a comprehensive database that encompasses over 35,000 content values for 500 different polyphenols in more than 400 foods. The database comprises a systematic collection of values reported in more than 1,300 scientific publications.

From a technological point of view, the potential use of anthocyanins as colorants in food and cosmetics has been extensively analyzed since they are the main hydrosoluble pigments from plant sources [97].

Anthocyanins are reactive molecules, highly susceptible to degradation. Their instability and the influence of concentration, pH, temperature, oxygen, light, and the presence of other compounds (phenolic acids, metals, enzymes) on color properties can limit anthocyanin use as food ingredients and additives. Likewise, anthocyanins may be degraded through several processes occurring during their extraction, processing and storage [48].

As it has been previously mentioned, the color of anthocyanins and its stability are strongly influenced by proton concentration (Figure 5). In solution, anthocyanin structures change from cationic (colored) to pseudobase (uncolored) forms. In acidic environments (pH 3 or lower), anthocyanins are present as flavylium cation and exhibit red to orange colors. When pH increases above 3, hydration of the flavylium cation takes place and a colorless carbinol pseudo-base is formed. The subsequent ring opening allows to the formation of an unstable chalcone pseudo-base [98].

The cationic form is stabilized at low pH values, which changes toward uncolored structures as the pH increases toward neutral values. According to Delgado-Vargas and Paredes-Lopez, [99] carbons hydroxylated at the positions 7 or 3′ favor quinonoidal equilibrium as the pH increases. Contrary to the pseudobase-uncolored forms, the quinonoidal structures are colored with hues tending to blue tincts.

Strawberry pulp represents a good source of anthocyanins that can be extracted by ethanol acidified with hydrochloric, tartaric, citric, lactic or phosphoric acid. In the work of de Vargas et al., [100] the authors evaluated different solvent volumes, numbers of extractions and contact times when separating pelargonidin 3-glucoside from the residue of strawberry processing. Four extractions during 12 min, using 20 mL of ethanol with 1% HCl, yielded a 98% extraction.

According to Wiczkowski et al., [101] fruits and vegetables are frequently subjected to several processing operations to obtain more suitable and attractive food products, as well as to achieve stability and longer shelf-life. In this case, anthocyanin stability is dependent on compound structure, plant matrix and process conditions, such as temperature and duration of the process. Environmental factors (i.e. oxygen, light, enzymes and microbial activity, associated substances, pH, etc.) affect the stability of anthocyanins. When studying the effect

of fermentation, storage and stewing on the content and composition of anthocyanins of red cabbage, Wiczkowski et al., [101] found that the applied treatments reduced anthocyanin content by 24%, 25% and 34% in fermented and stewed (30 or 60 min) red cabbage, respectively. Likewise, derivatives of cyanidin-3-diglucoside-5-glucoside acylated with sinapic acid showed the highest losses.
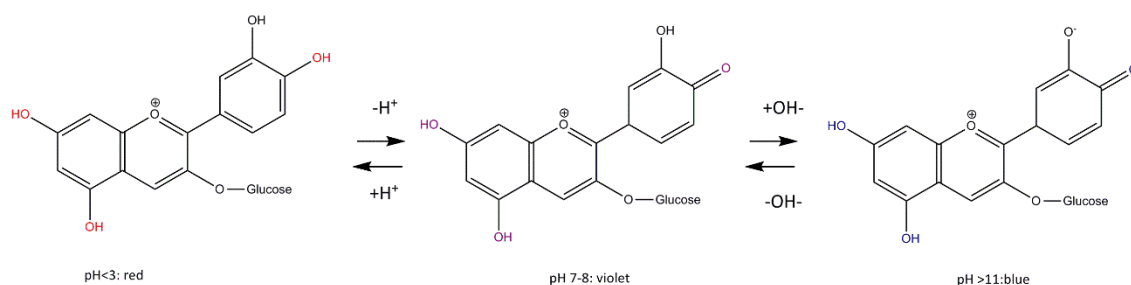

Figure 5. pH effects on anthocyanin chemical structure and color

Anthocyanin obtaining process from plant resources to be used as food ingredients requires extraction, concentration, and drying steps which expose anthocyanins to unfavorable environments. The latter can be conducted by simple thermal treatment, freeze drying, or spray drying. Spray drying has been demonstrated to be a particularly fast and gentle method for the drying of anthocyanin extracts [102].

The lower stability of anthocyanins compared to synthetic colorants has limited their use as pigments. However, anthocyanins are particularly interesting to the food industry due to their potentially beneficial health effects, which would represent the addition of a functional component to the food product, in addition to being more accepted by the consumers because of their natural origin.

Many approaches to increase anthocyanins stability have been studied, such as different encapsulation methods, copigmentation or chemical and enzymatic acylation.

Glycosyl acylation (esterification of the glycosyl groups with aliphatic or aromatic acids) has been related to increased anthocyanin stability against heat, light, pH changes, and digestive enzymes, and resistance to oxidation [103-105]. Acylation can reduce the sensitivity of anthocyanins to nucleophilic attack in aqueous medium by decreasing the molecule´s polarity (which is also related to anthocyanin increased resistance through the digestive system). Anthocyanin acylation also provides stability by steric hindrance effect and by the formation of zwitterions, which reduce the instability derived from pH increase [104]. Furthermore, the stacking effect of the acyl groups (mainly the aromatic ones) with the pyrylium ring of the anthocyanidin flavylium cations also enhances anthocyanin stability. Particularly, it has been stressed that acylation in the C_3 or C_5 positions of the anthocyanidin allows the formation of intramolecular copigmentation by π-π interactions with the benzopyrylium ring. Also, glycosyl groups with double bonds that may form a conjugated system with the benzene rings also confer photochemical stability to anthocyanins [104]. In this sense, anthocyanins extracted from red cabbage were the first to be commercialized and are highly appreciated in the food industry for being a natural product containing mostly diacylated anthocyanins which also exhibits high tinctorial strength [106]. Black carrot and red radish are also excellent sources of acylated anthocyanins with extracts available in a commercial form [106-107].

In the plant cells, anthocyanin sugar moieties are enzymatically acylated from acyl-CoA residues catalyzed by anthocyanin acyltransferases (AATs), whose cDNAs have been successfully expressed in *Escherichia coli* and yeasts [108]. By this approach, the biotechnological modification of plants may allow the synthesis of more stable anthocyanins. In the work of Suzuki et al., [109] cDNA coding for a 3-glucoside-specific malonyltransferase

from dahlia (*Dahlia variabilis*) was expressed in *E. coli* and the recombinant enzyme was introduced into petunia (*Petunia hybrida*) flowers, which led to the stabilization of the flower pigments. Nevertheless, when anthocyanins are sought for food uses, genetic modifications tend to be controversial and not well received by the consumers.

Anthocyanin artificial acylation has also been assayed by lipase-catalyzed transesterification using organic acids (especially cinnamic acids) as acyl donors. In the work of de Castro et al., [110] *Candida antarctica* lipase B was used to catalyze the acylation of polyphenol extracts from jabuticaba fruits (*Myrciaria cauliflora*) with palmitic acid as acyl donor. Furthermore, jambolan (*Syzygium cumini*) fruit anthocyanin transesterification with cinnamic acid was lipase-catalyzed with the same enzyme [111], and Yan et al. [112] assayed this lipase in the acylation of black rice extracts with aromatic carboxylic acids, which, in both cases, enhanced pigment thermostability and light resistivity. Cruz et al. [113] used a saturated fatty acid (stearic acid) as acyl donor and obtained more lipophilic derivatives which could potentially improve their bioavailability, despite exhibiting a lower antioxidant activity compared to their precursors.

Lipase transacylation represents an interesting way to increase anthocyanin stability; nevertheless, the lack of regiocontrol of the acylation in the sugar moiety is one of the issues to overcome for the broad application of this approach.

Encapsulation, complexation and spray-drying are other strategies that have been explored to attenuate anthocyanins degradation rate and have the advantage over enzymatic acylation of not modifying their natural structure.

Encapsulation proved to be an efficient method for the preservation and increase of the shelf-life of anthocyanins [114-118]. In this sense, Pereira Souza et al., [119] found that anthocyanins encapsulation in maltodextrin matrices, besides increasing the color stability, reduced the degradation of the pigments subjected to UV radiation. In the work of Guldiken et al. [120], liposomal entrapment in lecithin matrices also proved to be a shielding barrier for anthocyanins, protecting black carrot extracts against degradation by ascorbic acid. Particularly, it was stressed that the transparency of the liposomal system may allow its use in clear beverages [120].

The availability of a broad pool of biopolymers exhibiting different polarities and technological properties, many of them catalogued as GRAS, also allows for numerous formulations designed for specific uses. Furthermore, encapsulation using smart matrices designed for the controlled release of anthocyanins could help regulating the amount of pigments freed in a lapse of time in particular food systems or may allow to protect the ingested pigments when passing through the gastric system, thus increasing their bioavailability. In the work of Wang et al., [121], the inclusion of cellulose nanocrystals (CNC) in chitosan based microcapsules for anthocyanin encapsulation enhanced the rigidness and stability of the matrix compared to the addition of sodium tripolyphosphate (TPP). Both formulations exhibited similar release profiles at acidic pH, but the inclusion of CNC reduced the release percentage at neutral pH (7.4) compared to the TPP ones [121], which would be their expected behavior at intestinal conditions. Shi et al., [122] designed pH-sensitive microspheres of chitosan-coated TEMPO-oxidized Konjac glucomannan (OKGM), cross-linked via ferric ions (Fe^{3+}), which allowed for the simultaneous incorporation of hydrophobic (β-carotene) and hydrophilic (anthocyanins) antioxidants, their preservation in acidic gastric environment and the release of both antioxidants in simulated GI fluids, in addition to enhancing the thermostability of the antioxidant agents.

Amin et al., [123] used poly (lactide-co-glycolide) (PLGA) and polyethylene glycol (PEG)-2000 for the nanoencapsulation of anthocyanins, which allowed for increasing the attenuation of Alzheimer's, neuroinflammatory and neuroapoptotic markers in SH-SY5Y cell cultures. Authors also found that none of the assayed doses showed cytotoxic effect [123].

The combination of encapsulation methods with copigmentation proved to enhance the efficiency of anthocyanin preservation. An increased stability was observed for encapsulated black soybean anthocyanin in chitosan matrices copigmented with sinapic acid [124].

Chitosan and chondroitin sulfate-encapsulated elderberry anthocyanins copigmented with epigallocatechin gallate [125], and blackberry anthocyanins copigmented with rutin and ferulic acid and spray-dried with maltodextrin as polymeric solution have also been reported to present an increased stability [126]. It is worth to mention that the copigmentation with natural phenolic compounds of food origin has the advantage of not disabling the potential application of the encapsulated anthocyanins in the food, pharmaceutical and cosmetic industries. Furthermore, among the entrapping techniques explored to increase pigment stability, spray-drying of anthocyanins in aqueous biopolymeric solutions leads to obtaining powders with great solubility in water, which makes them particularly suitable for food applications [127-130]. Ratanapoompinyo et al., [131] evaluated the copigmentation of red cabbage anthocyanins with metal ions (Al^{3+}, Ca^{2+}, Fe^{3+}, and Sn^{2+}) and spray-dried with maltodextrin as feed solution, and the authors reported that ion addition accelerated anthocyanin degradation. Conversely, Luna-Vital et al., [132] found that the incorporation of alginate and Zn^{2+} ions to a commercial beverage added with purple corn anthocyanin-rich extract efficiently stabilized the anthocyanins against thermal or long-term storage conditions, compared to the alginate alone or the controls without alginate and Zn^{2+}.

Other approaches, such as complexation [133] and emulsification [134], are also promising techniques for increasing the resistance of anthocyanins to processing conditions. Nevertheless, despite the numerous advantages of anthocyanins as colorants (being natural products providing bright intense colors, and associated with health beneficial effects), the broad industrial use of anthocyanins as natural pigments is nowadays subject to the development of efficient stabilization methods to turn them into alternatives with competitiveness against synthetic dyes.

Final remarks
Epidemiologic and clinical studies (randomized controlled trials) provide increasing evidence of the benefits of fruit and vegetable consumption being related to the presence of bioactive compounds synthesized by plant cells and tissues. One of these natural plant products receiving growing attention due to their antioxidant and bioactive properties are anthocyanins; because they can prevent neurodegenerative disorders, diabetes, angio-cardio-pathologies, cancer, inflammation, and/or several deterioration processes related with aging. Added to their ability to prevent oxidation processes, anthocyanins can modulate key signaling pathways and gene regulation. Molecular targets of anthocyanins comprise to transporters and receptors, second messenger signaling molecules and kinase enzymes, transcription factors, promoters, growth factors, and defense enzymes.

Bioavailability (that includes the absorption, disposition, metabolism, and excretion of dietary components) has been a topic increasingly addressed in nutrition research. Formerly, anthocyanins were considered poorly bioavailable since low levels of phase II metabolites (i. e. compounds resulting from the introduction of reactive groups and the conjugation with polar compounds, mainly in the enterocytes and liver, forming sulfate, glucuronide, and/or methylated derivatives) were detected in the circulatory system and excreted in urine. However, epidemiologic observations led to consider that anthocyanins might either have a higher bioavailability than it was originally estimated or be very potent and highly active at nano- to micromolar plasmatic concentrations.

In vitro digestion models and microbial fermentation assays suggested that ingested anthocyanins are spontaneously or enzymatically degraded into phenolic derivatives, which are further metabolized, and these metabolites are present in the circulation for about 48 h

after ingestion. Recent evidence shows that anthocyanins are more bioavailable than previously perceived, when the diverse metabolites and catabolites are considered.

The interest on anthocyanin compounds is evidenced by the production of more than 6,500 publications since 2015, as shown by the results obtained for the search of the term "anthocyanins" in the main platforms of peer-reviewed scholarly literature. Although these phenolic substances show a broad range of biological activities, more research on safety and toxicological implications still needs to be conducted.

References

[1] Novel Media Nobel Prize in Chemistry 1915 - Presentation (accessed 25 Jun). Available from: http://www.nobelprize.org/nobel_prizes/chemistry/laureates/1915/present.html

[2] Robinson, R. Richard Willstätters Investigations of the Anthocyanins. *Naturwissenschaften*, **1932,** *20*(33), 612-618.

[3] Burton-Freeman, B.; Sandhu, A.; Edirisinghe, I. Chapter 35 - Anthocyanins A2 - In: *Nutraceuticals*; Gupta, Ramesh C., Ed.; Academic Press: Boston, **2016;** pp 489-500.

[4] Cronk, Q.; Ojeda, I. Bird-pollinated flowers in an evolutionary and molecular context. *J Exp Bot*, **2008,** *59*(4), 715-727.

[5] Santos, E.L.; Sales Maia, B.H.L.N.; Ferriani, A.P.; Dias Teixeira, S. Flavonoids: Classification, Biosynthesis and Chemical Ecology In: *Flavonoids - From Biosynthesis to Human Health*; Justino, D. J., Ed.; InTech: **2017**.

[6] Landi, M.; Tattini, M.; Gould, K.S. Multiple functional roles of anthocyanins in plant-environment interactions. *Environmental and Experimental Botany*, **2015,** *119*(Supplement C), 4-17.

[7] Gould, K.S. Nature's Swiss Army Knife: The Diverse Protective Roles of Anthocyanins in Leaves. *Journal of Biomedicine and Biotechnology*, **2004,** *2004*(5), 314-320.

[8] Li, P.; Li, Y.-J.; Zhang, F.-J.; Zhang, G.-Z.; Jiang, X.-Y.; Yu, H.-M.; Hou, B.-K. The Arabidopsis UDP-glycosyltransferases UGT79B2 and UGT79B3, contribute to cold, salt and drought stress tolerance via modulating anthocyanin accumulation. *The Plant Journal*, **2017,** *89*(1), 85-103.

[9] Kim, J.; Lee, W.J.; Vu, T.T.; Jeong, C.Y.; Hong, S.-W.; Lee, H. High accumulation of anthocyanins via the ectopic expression of AtDFR confers significant salt stress tolerance in Brassica napus L. *Plant Cell Reports*, **2017,** *36*(8), 1215-1224.

[10] Majetic, C.J.; Fetters, A.M.; Beck, O.M.; Stachnik, E.F.; Beam, K.M. Petunia floral trait plasticity in response to soil nitrogen content and subsequent impacts on insect visitation. *Flora*, **2017,** *232*(Supplement C), 183-193.

[11] Nicotra, A.B.; Atkin, O.K.; Bonser, S.P.; Davidson, A.M.; Finnegan, E.J.; Mathesius, U.; Poot, P.; Purugganan, M.D.; Richards, C.L.; Valladares, F.; van Kleunen, M. Plant phenotypic plasticity in a changing climate. *Trends in Plant Science*, **2010,** *15*(12), 684-692.

[12] Wang, L.-J.; Su, S.; Wu, J.; Du, H.; Li, S.-S.; Huo, J.-W.; Zhang, Y.; Wang, L.-S. Variation of anthocyanins and flavonols in Vaccinium uliginosum berry in Lesser Khingan Mountains and its antioxidant activity. *Food Chemistry*, **2014,** *160*(Supplement C), 357-364.

[13] Dar, T.A.; Uddin, M.; Ali, A.; Khan, M.M.A.; ul Hassan Dar, T. Understanding the Dynamics of Phosphorus Starvation and Plant Growth. In *Essential Plant Nutrients: Uptake, Use Efficiency, and Management*; Naeem, M.; Ansari, A. A.; Gill, S. S., Eds.; Springer International Publishing: Cham, **2017;** pp 147-154.

[14] Steyn, W.J.; Wand, S.J.E.; Holcroft, D.M.; Jacobs, G. Anthocyanins in vegetative tissues: a proposed unified function in photoprotection. *New Phytologist*, **2002,** *155*(3), 349-361.

[15] Chiou, A.; Panagopoulou, E.A.; Gatzali, F.; De Marchi, S.; Karathanos, V.T. Anthocyanins content and antioxidant capacity of Corinthian currants (Vitis vinifera L., var. Apyrena). *Food Chemistry*, **2014,** *146*(Supplement C), 157-165.

[16] Abad-García, B.; Berrueta, L.A.; Garmón-Lobato, S.; Gallo, B.; Vicente, F. A general analytical strategy for the characterization of phenolic compounds in fruit juices by high-

performance liquid chromatography with diode array detection coupled to electrospray ionization and triple quadrupole mass spectrometry. *Journal of Chromatography A*, **2009,** *1216*(28), 5398-5415.

[17] Stalmach, A. Chapter 42 - Bioavailability of Dietary Anthocyanins and Hydroxycinnamic Acids In: *Polyphenols in Human Health and Disease*; Watson, R.R.; Preedy, V.R.; Zibadi, S., Eds.; Academic Press: San Diego, **2014;** pp 561-576.

[18] He, J.; Giusti, M.M. Anthocyanins: Natural Colorants with Health-Promoting Properties. *Annual Review of Food Science and Technology*, **2010,** *1*(1), 163-187.

[19] Jamar, G.; Estadella, D.; Pisani, L.P. Contribution of anthocyanin-rich foods in obesity control through gut microbiota interactions. *BioFactors*, **2017,** *43*(4), 507-516.

[20] Ali, H.M.; Almagribi, W.; Al-Rashidi, M.N. Antiradical and reductant activities of anthocyanidins and anthocyanins, structure–activity relationship and synthesis. *Food Chemistry*, **2016,** *194*(Supplement C), 1275-1282.

[21] Chaves, V.C.; Calvete, E.; Reginatto, F.H. Quality properties and antioxidant activity of seven strawberry (Fragaria x ananassa duch) cultivars. *Scientia Horticulturae*, **2017,** *225*(Supplement C), 293-298.

[22] Thilavech, T.; Abeywardena, M.Y.; Adams, M.; Dallimore, J.; Adisakwattana, S. Naturally occurring anthocyanin cyanidin-3-rutinoside possesses inherent vasorelaxant actions and prevents methylglyoxal-induced vascular dysfunction in rat aorta and mesenteric arterial bed. *Biomedicine & Pharmacotherapy*, **2017,** *95*(Supplement C), 1251-1259.

[23] Bowsher, C.; Steer, M.W.; Tobin, A.K. *Plant Biochemistry*. Garland Science: New York, **2008**.

[24] Falcone Ferreyra, M.L.; Rius, S.P.; Casati, P. Flavonoids: biosynthesis, biological functions, and biotechnological applications. *Frontiers in Plant Science*, **2012,** *3*, 222.

[25] Jain, G.; Gould, K.S. Are betalain pigments the functional homologues of anthocyanins in plants? *Environmental and Experimental Botany*, **2015,** *119*(Supplement C), 48-53.

[26] Miguel, M.G. Anthocyanins: Antioxidant and/or anti-inflammatory activities. *Journal of Applied Pharmaceutical Science*, **2011,** *1*(6), 7-15.

[27] Tsuda, T. Recent progress in anti-obesity and anti-diabetes effect of berries. *Antioxidants*, **2016,** *5*(2), 13.

[28] Lee, S.G.; Kim, B.; Yang, Y.; Pham, T.X.; Park, Y.-K.; Manatou, J.; Koo, S.I.; Chun, O.K.; Lee, J.-Y. Berry anthocyanins suppress the expression and secretion of proinflammatory mediators in macrophages by inhibiting nuclear translocation of NF-κB independent of NRF2-mediated mechanism. *The Journal of Nutritional Biochemistry*, **2014,** *25*(4), 404-411.

[29] Wu, T.; Tang, Q.; Yu, Z.; Gao, Z.; Hu, H.; Chen, W.; Zheng, X.; Yu, T. Inhibitory effects of sweet cherry anthocyanins on the obesity development in C57BL/6 mice. *International Journal of Food Sciences and Nutrition*, **2014,** *65*(3), 351-359.

[30] Noratto, G.D.; Angel-Morales, G.; Talcott, S.T.; Mertens-Talcott, S.U. Polyphenolics from Açaí (Euterpe oleracea Mart.) and Red Muscadine Grape (Vitis rotundifolia) Protect Human Umbilical Vascular Endothelial Cells (HUVEC) from Glucose- and Lipopolysaccharide (LPS)-Induced Inflammation and Target MicroRNA-126. *J Agric Food Chem*, **2011,** *59*(14), 7999-8012.

[31] Speciale, A.; Virgili, F.; Saija, A.; Cimino, F. Chapter 72 - Anthocyanins in Vascular Diseases In: *Polyphenols in Human Health and Disease*; Watson, R.R.; Preedy, V.R.; Zibadi, S., Eds.; Academic Press: San Diego, **2014;** pp 923-941.

[32] Zhang, H.; Hassan, Y.I.; Renaud, J.; Liu, R.; Yang, C.; Sun, Y.; Tsao, R. Bioaccessibility, bioavailability, and anti-inflammatory effects of anthocyanins from purple root vegetables using mono- and co-culture cell models. *Molecular Nutrition & Food Research*, **2017,** *61*(10), 1600928.

[33] Venancio, V.P.; Cipriano, P.A.; Kim, H.; Antunes, L.M.G.; Talcott, S.T.; Mertens-Talcott, S.U. Cocoplum (Chrysobalanus icaco L.) anthocyanins exert anti-inflammatory activity in human colon cancer and non-malignant colon cells. *Food & Function*, **2017,** *8*(1), 307-314.

[34] Alvarez-Suarez, J.M.; Carrillo-Perdomo, E.; Aller, A.; Giampieri, F.; Gasparrini, M.; González-Pérez, L.; Beltrán-Ayala, P.; Battino, M. Anti-inflammatory effect of Capuli cherry against LPS-induced cytotoxic damage in RAW 264.7 macrophages. *Food and Chemical Toxicology*, **2017,** *102*(Supplement C), 46-52.

[35] Winter, A.N.; Brenner, M.C.; Punessen, N.; Snodgrass, M.; Byars, C.; Arora, Y.; Linseman, D.A. Comparison of the neuroprotective and anti-inflammatory effects of the anthocyanin metabolites, protocatechuic acid and 4-Hydroxybenzoic acid. *Oxidative Medicine and Cellular Longevity*, **2017,** *2017*, 13.

[36] Oghumu, S.; Knobloch, T.; Ahn-Jarvis, J.; Weghorst, L.; Horvath, K.; Geuy, P.; Weghorst, C. Abstract 5264: Chemoprevention of rat oral carcinogenesis by black raspberry phytochemicals. *Cancer Research*, **2017,** *77*(13 Supplement), 5264-5264.

[37] Lin, B.-W.; Gong, C.-C.; Song, H.-F.; Cui, Y.-Y. Effects of anthocyanins on the prevention and treatment of cancer. *British Journal of Pharmacology*, **2017,** *174*(11), 1226-1243.

[38] Kuntz, S.; Kunz, C.; Rudloff, S. Inhibition of pancreatic cancer cell migration by plasma anthocyanins isolated from healthy volunteers receiving an anthocyanin-rich berry juice. *European Journal of Nutrition*, **2017,** *56*(1), 203-214.

[39] Lippert, E.; Ruemmele, P.; Obermeier, F.; Goelder, S.; Kunst, C.; Rogler, G.; Dunger, N.; Messmann, H.; Hartmann, A.; Endlicher, E. Anthocyanins Prevent Colorectal Cancer Development in a Mouse Model. *Digestion*, **2017,** *95*(4), 275-280.

[40] Wang, L.-S.; Hecht, S.S.; Carmella, S.G.; Yu, N.; Larue, B.; Henry, C.; McIntyre, C.; Rocha, C.; Lechner, J.F.; Stoner, G.D. Anthocyanins in black raspberries prevent esophageal tumors in rats. *Cancer Prevention Research*, **2009,** *2*(1), 84-93.

[41] Mazewski, C.; Liang, K.; Gonzalez de Mejia, E. Inhibitory potential of anthocyanin-rich purple and red corn extracts on human colorectal cancer cell proliferation in vitro. *Journal of Functional Foods*, **2017,** *34*(Supplement C), 254-265.

[42] Limsitthichaikoon, S.; Priprem, A.; Damrongrungruang, T.; Limphirat, W.; Kukhetpitakwong, R. Improvement of chemical stability and activities of anthocyanins by complexation. *Current Bioactive Compounds*, **2016,** *12*(1), 17-24.

[43] Priprem, A.; Limsitthichaikoon, S.; Sukkhamduang, W.; Limphirat, W.; Thapphasaraphong, S.; Nualkaew, N. Anthocyanin complex improves stability with *in vitro* localized UVA protective effect. *Current Bioactive Compounds*, **2017,** *13*(4), 333-339.

[44] Cassidy, A. Berry anthocyanin intake and cardiovascular health. *Molecular Aspects of Medicine*, **2017,** 61, 76-82.

[45] Bobinaitė, R.; Viškelis, J. Anthocyanins: occurrence, bioactivity and bioavailability, with special reference to the anthocyanins of raspberries (a review). *Sodininkystė ir Daržininkystė*, **2013,** *32*(1/2), 39-47.

[46] Bhaswant, M.; Shafie, S.R.; Mathai, M.L.; Mouatt, P.; Brown, L. Anthocyanins in chokeberry and purple maize attenuate diet-induced metabolic syndrome in rats. *Nutrition*, **2017,** *41*(Supplement C), 24-31.

[47] Jennings, A.; Welch, A.A.; Spector, T.; Macgregor, A.; Cassidy, A. Intakes of Anthocyanins and Flavones Are Associated with Biomarkers of Insulin Resistance and Inflammation in Women. *The Journal of Nutrition*, **2014,** *144*(2), 202-208.

[48] Fernandes, I.; Faria, A.; Calhau, C.; de Freitas, V.; Mateus, N. Bioavailability of anthocyanins and derivatives. *Journal of Functional Foods*, **2014,** *7*(Supplement C), 54-66.

[49] Scholz; Williamson Interactions affecting the bioavailability of dietary polyphenols *in vivo*. *International Journal for Vitamin and Nutrition Research*, **2007,** *77*(3), 224-235.

[50] Goszcz, K.; Deakin, S.J.; Duthie, G.G.; Stewart, D.; Megson, I.L. Bioavailable concentrations of delphinidin and its metabolite, gallic acid, induce antioxidant protection associated with increased intracellular glutathione in cultured endothelial cells. *Oxidative Medicine and Cellular Longevity*, **2017,** *2017*, 17.

[51] Talavéra, S.; Felgines, C.; Texier, O.; Besson, C.; Lamaison, J.-L.; Rémésy, C. Anthocyanins are efficiently absorbed from the stomach in anesthetized rats. *The Journal of Nutrition*, **2003,** *133*(12), 4178-4182.

[52] Atnip, A.A.; Sigurdson, G.T.; Bomser, J.; Giusti, M.M. Time, concentration, and pH-dependent transport and uptake of anthocyanins in a human gastric epithelial (NCI-N87) cell line. *International Journal of Molecular Sciences*, **2017,** *18*(2), 446.

[53] Matuschek, M.C.; Hendriks, W.H.; McGhie, T.K.; Reynolds, G.W. The jejunum is the main site of absorption for anthocyanins in mice. *The Journal of Nutritional Biochemistry*, **2006,** *17*(1), 31-36.

[54] Mueller, D.; Jung, K.; Winter, M.; Rogoll, D.; Melcher, R.; Richling, E. Human intervention study to investigate the intestinal accessibility and bioavailability of anthocyanins from bilberries. *Food Chemistry*, **2017,** *231*(Supplement C), 275-286.

[55] Pan, P.; Lam, V.; Salzman, N.; Huang, Y.-W.; Yu, J.; Zhang, J.; Wang, L.-S. Black Raspberries and Their Anthocyanin and Fiber Fractions Alter the Composition and Diversity of Gut Microbiota in F-344 Rats. *Nutrition and Cancer*, **2017,** *69*(6), 943-951.

[56] Tsuda, T. Dietary anthocyanin-rich plants: Biochemical basis and recent progress in health benefits studies. *Molecular Nutrition & Food Research*, **2012,** *56*(1), 159-170.

[57] Leong, S.Y.; Burritt, D.J.; Hocquel, A.; Penberthy, A.; Oey, I. The relationship between the anthocyanin and vitamin C contents of red-fleshed sweet cherries and the ability of fruit digests to reduce hydrogen peroxide-induced oxidative stress in Caco-2 cells. *Food Chemistry*, **2017,** *227*(Supplement C), 404-412.

[58] Mazza, G.; Miniati, E. *Anthocyanins in fruits, vegetables, and grains*. CRC Press: Boca Raton, Florida, **1993**.

[59] da Costa, C.T.; Horton, D.; Margolis, S.A. Analysis of anthocyanins in foods by liquid chromatography, liquid chromatography–mass spectrometry and capillary electrophoresis. *Journal of Chromatography A*, **2000,** *881*(1), 403-410.

[60] Cacace, J.E.; Mazza, G. Optimization of Extraction of Anthocyanins from Black Currants with Aqueous Ethanol. *Journal of Food Science*, **2003,** *68*(1), 240-248.

[61] Paunović, S.M.; Mašković, P.; Nikolić, M.; Miletić, R. Bioactive compounds and antimicrobial activity of black currant (Ribes nigrum L.) berries and leaves extract obtained by different soil management system. *Scientia Horticulturae*, **2017,** *222*(Supplement C), 69-75.

[62] Veberic, R.; Jakopic, J.; Stampar, F.; Schmitzer, V. European elderberry (*Sambucus nigra* L.) rich in sugars, organic acids, anthocyanins and selected polyphenols. *Food Chemistry*, **2009,** *114*(2), 511-515.

[63] Lee, J.; Dossett, M.; Finn, C.E. Rubus fruit phenolic research: The good, the bad, and the confusing. *Food Chemistry*, **2012,** *130*(4), 785-796.

[64] Colak, N.; Primetta, A.K.; Riihinen, K.R.; Jaakola, L.; Grúz, J.; Strnad, M.; Torun, H.; Ayaz, F.A. Phenolic compounds and antioxidant capacity in different-colored and non-pigmented berries of bilberry (Vaccinium myrtillus L.). *Food Bioscience*, **2017,** *20*(Supplement C), 67-78.

[65] Tolić, M.-T.; Jurčević, I.L.; Krbavčić, I.P.; Marković, K.; Vahčić, N. Phenolic Content, Antioxidant Capacity and Quality of Chokeberry (Aronia melanocarpa) Products. *Food Technology and Biotechnology*, **2015,** *53*(2), 171-179.

[66] Cesa, S.; Carradori, S.; Bellagamba, G.; Locatelli, M.; Casadei, M.A.; Masci, A.; Paolicelli, P. Evaluation of processing effects on anthocyanin content and colour modifications of blueberry (Vaccinium spp.) extracts: Comparison between HPLC-DAD and CIELAB analyses. *Food Chemistry*, **2017,** *232*(Supplement C), 114-123.

[67] de Cássia Rodrigues Batista, C.; de Oliveira, M.S.; Araújo, M.E.; Rodrigues, A.M.C.; Botelho, J.R.S.; da Silva Souza Filho, A.P.; Machado, N.T.; Carvalho, R.N. Supercritical CO2 extraction of açaí (*Euterpe oleracea*) berry oil: Global yield, fatty acids, allelopathic activities, and determination of phenolic and anthocyanins total compounds in the residual pulp. *The Journal of Supercritical Fluids*, **2016,** *107*(Supplement C), 364-369.

[68] Rumalla, C.S.; Avula, B.; Wang, Y.H.; Smillie, T.J.; Khan, I.A. Method Development and Analysis of Anthocyanins from Acai (*Euterpe oleracea Mart*.) Berries and Dietary Supplements using HPTLC. *Planta Med*, **2011,** *77*(05), 70.

[69] Schulz, M.; da Silva Campelo Borges, G.; Gonzaga, L.V.; Oliveira Costa, A.C.; Fett, R. Juçara fruit (*Euterpe edulis Mart.*): Sustainable exploitation of a source of bioactive compounds. *Food Research International*, **2016,** *89*(Part 1), 14-26.

[70] Ludwig, I.A.; Mena, P.; Calani, L.; Borges, G.; Pereira-Caro, G.; Bresciani, L.; Del Rio, D.; Lean, M.E.J.; Crozier, A. New insights into the bioavailability of red raspberry anthocyanins and ellagitannins. *Free Radical Biology and Medicine*, **2015,** *89*(Supplement C), 758-769.

[71] Teng, H.; Fang, T.; Lin, Q.; Song, H.; Liu, B.; Chen, L. Red raspberry and its anthocyanins: Bioactivity beyond antioxidant capacity. *Trends in Food Science & Technology*, **2017,** *66*(Supplement C), 153-165.

[72] Wiesenborn, D.P.; Xu, M.; Chang, K.C.; Schwarz, J.G. Pigment removal and pectin loss during the continuous, countercurrent washing of sunflower heads. *Transactions of the American Society of Agricultural Engineers*, **1996,** *39*(5), 1781-1787.

[73] Faramarzi, S.; Pacifico, S.; Yadollahi, A.; Lettieri, A.; Nocera, P.; Piccolella, S. Red-fleshed Apples: Old Autochthonous Fruits as a Novel Source of Anthocyanin Antioxidants. *Plant Foods for Human Nutrition*, **2015,** *70*(3), 324-330.

[74] Mazza, G.; Francis, F.J. Anthocyanins in grapes and grape products. *Critical Reviews in Food Science and Nutrition*, **1995,** *35*(4), 341-371.

[75] Giusti, M.M.; Rodríguez-Saona, L.E.; Baggett, J.R.; Reed, G.L.; Durst, R.W.; Wrolstad, R.E. Anthocyanin pigment composition of red radish cultivars as potential food colorants. *Journal of Food Science*, **1998,** *63*(2), 219-224.

[76] Niño-Medina, G.; Urías-Orona, V.; Muy-Rangel, M.D.; Heredia, J.B. Structure and content of phenolics in eggplant (*Solanum melongena*) - a review. *South African Journal of Botany*, **2017,** *111*(Supplement C), 161-169.

[77] Zaro, M.J.; Keunchkarian, S.; Chaves, A.R.; Vicente, A.R.; Concellón, A. Changes in bioactive compounds and response to postharvest storage conditions in purple eggplants as affected by fruit developmental stage. *Postharvest Biology and Technology*, **2014,** *96*(Supplement C), 110-117.

[78] Ahmadiani, N.; Robbins, R.J.; Collins, T.M.; Giusti, M.M. Anthocyanins contents, profiles, and color characteristics of red cabbage extracts from different cultivars and maturity stages. *J Agric Food Chem*, **2014,** *62*(30), 7524-7531.

[79] Usenik, V.; Štampar, F.; Veberič, R. Anthocyanins and fruit colour in plums (*Prunus domestica L.*) during ripening. *Food Chemistry*, **2009,** *114*(2), 529-534.

[80] Lopes da Silva, F.; Escribano-Bailón, M.T.; Pérez Alonso, J.J.; Rivas-Gonzalo, J.C.; Santos-Buelga, C. Anthocyanin pigments in strawberry. *LWT - Food Science and Technology*, **2007,** *40*(2), 374-382.

[81] Ivanova-Petropulos, V.; Hermosín-Gutiérrez, I.; Boros, B.; Stefova, M.; Stafilov, T.; Vojnoski, B.; Dörnyei, Á.; Kilár, F. Phenolic compounds and antioxidant activity of Macedonian red wines. *Journal of Food Composition and Analysis*, **2015,** *41*(Supplement C), 1-14.

[82] Romero, C.; Brenes, M.; García, P.; García, A.; Garrido, A. Polyphenol Changes during Fermentation of Naturally Black Olives. *J Agric Food Chem*, **2004,** *52*(7), 1973-1979.

[83] Goulas, V.; Charisiadis, P.; Gerothanassis, I.P.; Manganaris, G.A. Classification, Biotransformation and Antioxidant Activity of Olive Fruit Biophenols: A Review. *Current Bioactive Compounds*, **2012,** *8*(3), 232-239.

[84] Moro, L.; Hassimotto, N.M.A.; Purgatto, E. Postharvest Auxin and Methyl Jasmonate Effect on Anthocyanin Biosynthesis in Red Raspberry (Rubus idaeus L.). *Journal of Plant Growth Regulation*, **2017,** *36*(3), 773-782.

[85] Ferreyra, R.M.; Viña, S.Z.; Mugridge, A.; Chaves, A.R. Growth and ripening season effects on antioxidant capacity of strawberry cultivar Selva. *Scientia Horticulturae*, **2007,** *112*(1), 27-32.

[86] Zhu, Y.-f.; Su, J.; Yao, G.-f.; Liu, H.-n.; Gu, C.; Qin, M.-f.; Bai, B.; Cai, S.-s.; Wang, G.-m.; Wang, R.-z.; Shu, Q.; Wu, J. Different light-response patterns of coloration and related gene expression in red pears (Pyrus L.). *Scientia Horticulturae*, **2018,** *229*(Supplement C), 240-251.

[87] Giribaldi, M.; Gény, L.; Delrot, S.; Schubert, A. Proteomic analysis of the effects of ABA treatments on ripening Vitis vinifera berries. *J Exp Bot*, **2010,** *61*(9), 2447-2458.

[88] Koyama, K.; Sadamatsu, K.; Goto-Yamamoto, N. Abscisic acid stimulated ripening and gene expression in berry skins of the Cabernet Sauvignon grape. *Functional & Integrative Genomics*, **2010,** *10*(3), 367-381.

[89] Oh, H.D.; Yu, D.J.; Chung, S.W.; Chea, S.; Lee, H.J. Abscisic acid stimulates anthocyanin accumulation in 'Jersey' highbush blueberry fruits during ripening. *Food Chemistry*, **2018,** *244*(Supplement C), 403-407.

[90] Jiang, Y.; Joyce, D.C. ABA effects on ethylene production, PAL activity, anthocyanin and phenolic contents of strawberry fruit. *Plant Growth Regulation*, **2003,** *39*(2), 171-174.

[91] Sousa, C.; Mateus, N.; Silva, A.M.S.; González-Paramás, A.M.; Santos-Buelga, C.; Freitas, V.d. Structural and chromatic characterization of a new Malvidin 3-glucoside–vanillyl–catechin pigment. *Food Chemistry*, **2007,** *102*(4), 1344-1351.

[92] Moreno-Arribas, M.V.; Polo, C. *Wine Chemistry and Biochemistry*. Springer New York: **2008**.

[93] Escribano-Bailón, M.T.; Santos-Buelga, C.; Rivas-Gonzalo, J.C. Anthocyanins in cereals. *Journal of Chromatography A*, **2004,** *1054*(1), 129-141.

[94] Su, X.; Rhodes, D.; wang, w.; Yu, M.; Katz, B.; Tomich, J. Phenotypic diversity of anthocyanins in 25 sorghum accessions. *The FASEB Journal*, **2015,** *29*(1 Supplement).

[95] Neveu, V.; Perez-Jiménez, J.; Vos, F.; Crespy, V.; du Chaffaut, L.; Mennen, L.; Knox, C.; Eisner, R.; Cruz, J.; Wishart, D.; Scalbert, A. Phenol-Explorer: an online comprehensive database on polyphenol contents in foods. *Database: The Journal of Biological Databases and Curation*, **2010,** *2010*, bap024.

[96] Rothwell, J.A.; Perez-Jimenez, J.; Neveu, V.; Medina-Remón, A.; M'Hiri, N.; García-Lobato, P.; Manach, C.; Knox, C.; Eisner, R.; Wishart, D.S.; Scalbert, A. Phenol-Explorer 3.0: a major update of the Phenol-Explorer database to incorporate data on the effects of food processing on polyphenol content. *Database: The Journal of Biological Databases and Curation*, **2013,** *2013*, bat070.

[97] Grajeda-Iglesias, C.; Salas, E.; Barouh, N.; Baréa, B.; Figueroa-Espinoza, M.C. Lipophilization and MS characterization of the main anthocyanins purified from hibiscus flowers. *Food Chemistry*, **2017,** *230*(Supplement C), 189-194.

[98] Howard, L.R.; Brownmiller, C.; Mauromoustakos, A.; Prior, R.L. Improved stability of blueberry juice anthocyanins by acidification and refrigeration. *Journal of Berry Research*, **2016,** *6*(2), 189-201.

[99] Delgado-Vargas, F.; Paredes-Lopez, O. *Natural Colorants for Food and Nutraceutical Uses*. CRC Press: Boca Ratón, **2002**.

[100] de Vargas, E.F.; Jablonsky, A.; Flores, S.H.; de Oliveira Rios, A. Pelargonidin 3-Glucoside Extraction from the Residue from Strawberry Processing (Fragaria X Ananassa). *Current Bioactive Compounds*, **2016,** *12*(4), 269-275.

[101] Wiczkowski, W.; Szawara-Nowak, D.; Topolska, J. Changes in the content and composition of anthocyanins in red cabbage and its antioxidant capacity during fermentation, storage and stewing. *Food Chemistry*, **2015,** *167*(Supplement C), 115-123.

[102] Robert, P.; Fredes, C. The Encapsulation of Anthocyanins from Berry-Type Fruits. Trends in Foods. *Molecules*, **2015,** *20*(4), 5875.

[103] Honda, T.; Saito, N. Recent Progress in the Chemistry of Polyacylated Anthocyanins as Flower Color Pigments. *Heterocycles*, **2002,** *56*(1-2), 633-692.

[104] Zhao, C.-L.; Yu, Y.-Q.; Chen, Z.-J.; Wen, G.-S.; Wei, F.-G.; Zheng, Q.; Wang, C.-D.; Xiao, X.-L. Stability-increasing effects of anthocyanin glycosyl acylation. *Food Chemistry*, **2017,** *214*(Supplement C), 119-128.

[105] Giusti, M.M.; Wrolstad, R.E. Acylated anthocyanins from edible sources and their applications in food systems. *Biochemical Engineering Journal*, **2003,** *14*(3), 217-225.

[106] Bakowska-Barczak, A. Acylated anthocyanins as stable, natural food colorants – a review. *Polish Journal of Food and Nutrition Sciences*, **2005,** *14/55*(2), 107–116.

[107] Malien-Aubert, C.; Dangles, O.; Amiot, M.J. Color Stability of Commercial Anthocyanin-Based Extracts in Relation to the Phenolic Composition. Protective Effects by Intra- and Intermolecular Copigmentation. *J Agric Food Chem*, **2001,** *49*(1), 170-176.

[108] Nakayama, T.; Suzuki, H.; Nishino, T. Anthocyanin acyltransferases: specificities, mechanism, phylogenetics, and applications. *Journal of Molecular Catalysis B: Enzymatic*, **2003,** *23*(2), 117-132.

[109] Suzuki, H.; Nakayama, T.; Yonekura-Sakakibara, K.; Fukui, Y.; Nakamura, N.; Yamaguchi, M.-a.; Tanaka, Y.; Kusumi, T.; Nishino, T. cDNA Cloning, Heterologous Expressions, and Functional Characterization of Malonyl-Coenzyme A:Anthocyanidin 3-O-Glucoside-6"-O-Malonyltransferase from Dahlia Flowers. *Plant Physiology*, **2002,** *130*(4), 2142-2151.

[110] de Castro, V.C.; da Silva, P.H.A.; de Oliveira, E.B.; Desobry, S.; Humeau, C. Extraction, identification and enzymatic synthesis of acylated derivatives of anthocyanins from jaboticaba (Myrciaria cauliflora) fruits. *International Journal of Food Science & Technology*, **2014,** *49*(1), 196-204.

[111] Sari, P.; Setiawan, A.; Siswoyo, T.A. Stability and antioxidant activity of acylated jambolan (*Syzygium cumini*) anthocyanins synthesized by lipase-catalyzed transesterification. *International Food Research Journal*, **2015,** *22*(2), 671-676.

[112] Yan, Z.; Li, C.; Zhang, L.; Liu, Q.; Ou, S.; Zeng, X. Enzymatic acylation of anthocyanin isolated from black rice with methyl aromatic acid ester as donor: stability of the acylated derivatives. *J Agric Food Chem*, **2016,** *64*(5), 1137-1143.

[113] Cruz, L.; Fernandes, V.C.; Araújo, P.; Mateus, N.; de Freitas, V. Synthesis, characterisation and antioxidant features of procyanidin B4 and malvidin-3-glucoside stearic acid derivatives. *Food Chemistry*, **2015,** *174*(Supplement C), 480-486.

[114] Shaddel, R.; Hesari, J.; Azadmard-Damirchi, S.; Hamishehkar, H.; Fathi-Achachlouei, B.; Huang, Q. Use of gelatin and gum Arabic for encapsulation of black raspberry anthocyanins by complex coacervation. *International Journal of Biological Macromolecules*, **2017,** 107 (Part B), 1800-1810.

[115] de Moura, S.C.S.R.; Berling, C.L.; Germer, S.P.M.; Alvim, I.D.; Hubinger, M.D. Encapsulating anthocyanins from *Hibiscus sabdariffa L*. calyces by ionic gelation: Pigment stability during storage of microparticles. *Food Chemistry*, **2018,** *241*(Supplement C), 317-327.

[116] Ge, J.; Yue, P.; Chi, J.; Liang, J.; Gao, X. Formation and stability of anthocyanins-loaded nanocomplexes prepared with chitosan hydrochloride and carboxymethyl chitosan. *Food Hydrocolloids*, **2018,** *74*(Supplement C), 23-31.

[117] He, B.; Ge, J.; Yue, P.; Yue, X.; Fu, R.; Liang, J.; Gao, X. Loading of anthocyanins on chitosan nanoparticles influences anthocyanin degradation in gastrointestinal fluids and stability in a beverage. *Food Chemistry*, **2017,** *221*(Supplement C), 1671-1677.

[118] Moser, P.; Telis, V.R.N.; de Andrade Neves, N.; García-Romero, E.; Gómez-Alonso, S.; Hermosín-Gutiérrez, I. Storage stability of phenolic compounds in powdered BRS Violeta grape juice microencapsulated with protein and maltodextrin blends. *Food Chemistry*, **2017,** *214*(Supplement C), 308-318.

[119] Pereira Souza, A.C.; Deyse Gurak, P.; Damasceno Ferreira Marczak, L. Maltodextrin, pectin and soy protein isolate as carrier agents in the encapsulation of anthocyanins-rich extract from jaboticaba pomace. *Food and Bioproducts Processing*, **2017,** *102*(Supplement C), 186-194.

[120] Guldiken, B.; Gibis, M.; Boyacioglu, D.; Capanoglu, E.; Weiss, J. Impact of liposomal encapsulation on degradation of anthocyanins of black carrot extract by adding ascorbic acid. *Food & Function*, **2017,** *8*(3), 1085-1093.

[121] Wang, W.; Jung, J.; Zhao, Y. Chitosan-cellulose nanocrystal microencapsulation to improve encapsulation efficiency and stability of entrapped fruit anthocyanins. *Carbohydrate Polymers*, **2017,** *157*(Supplement C), 1246-1253.

[122] Shi, M.; Bai, J.; Zhao, L.; Yu, X.; Liang, J.; Liu, Y.; Nord, W.; Li, Y. Co-loading and intestine-specific delivery of multiple antioxidants in pH-responsive microspheres based on TEMPO-oxidized polysaccharides. *Carbohydrate Polymers*, **2017,** *157*(Supplement C), 858-865.

[123] Amin, F.U.; Shah, S.A.; Badshah, H.; Khan, M.; Kim, M.O. Anthocyanins encapsulated by PLGA@PEG nanoparticles potentially improved its free radical scavenging capabilities via p38/JNK pathway against Aβ1–42-induced oxidative stress. *Journal of Nanobiotechnology*, **2017,** *15*(1), 12.

[124] Ko, A.; Lee, J.-S.; Sop Nam, H.; Gyu Lee, H. Stabilization of black soybean anthocyanin by chitosan nanoencapsulation and copigmentation. *Journal of Food Biochemistry*, **2017,** *41*(2), e12316-n/a.

[125] Tan, C.; Selig, M.J.; Abbaspourrad, A. Anthocyanin stabilization by chitosan-chondroitin sulfate polyelectrolyte complexation integrating catechin co-pigmentation. *Carbohydrate Polymers*, **2018,** *181*(Supplement C), 124-131.

[126] Weber, F.; Boch, K.; Schieber, A. Influence of copigmentation on the stability of spray dried anthocyanins from blackberry. *LWT - Food Science and Technology*, **2017,** *75*(Supplement C), 72-77.

[127] Yingngam, B.; Tantiraksaroj, K.; Taweetao, T.; Rungseevijitprapa, W.; Supaka, N.; Brantner, A.H. Modeling and stability study of the anthocyanin-rich maoberry fruit extract in the fast-dissolving spray-dried microparticles. *Powder Technology*, **2018,** *325*(Supplement C), 261-270.

[128] Ortiz-Basurto, R.I.; Rubio-Ibarra, M.E.; Ragazzo-Sanchez, J.A.; Beristain, C.I.; Jiménez-Fernández, M. Microencapsulation of Eugenia uniflora L. juice by spray drying using fructans with different degrees of polymerisation. *Carbohydrate Polymers*, **2017,** *175*(Supplement C), 603-609.

[129] Kalušević, A.; Lević, S.; Čalija, B.; Pantić, M.; Belović, M.; Pavlović, V.; Bugarski, B.; Milić, J.; Žilić, S.; Nedović, V. Microencapsulation of anthocyanin-rich black soybean coat extract by spray drying using maltodextrin, gum Arabic and skimmed milk powder. *Journal of Microencapsulation*, **2017,** *34*(5), 475-487.

[130] Begum, Y.A.; Deka, S.C. Stability of spray-dried microencapsulated anthocyanins extracted from culinary banana bract. *International Journal of Food Properties*, **2017,** *20*(12), 3135-3148.

[131] Ratanapoompinyo, J.; Nguyen, L.T.; Devkota, L.; Shrestha, P. The effects of selected metal ions on the stability of red cabbage anthocyanins and total phenolic compounds subjected to encapsulation process. *Journal of Food Processing and Preservation*, **2017,** *41*(6), e13234-n/a.

[132] Luna-Vital, D.; Cortez, R.; Ongkowijoyo, P.; Gonzalez de Mejia, E. Protection of color and chemical degradation of anthocyanin from purple corn (Zea mays L.) by zinc ions and alginate through chemical interaction in a beverage model. *Food Research International*, **2018,** *105*(Supplement C), 169-177.

[133] Fernandes, A.; Rocha, M.A.A.; Santos, L.M.N.B.F.; Brás, J.; Oliveira, J.; Mateus, N.; de Freitas, V. Blackberry anthocyanins: β-Cyclodextrin fortification for thermal and gastrointestinal stabilization. *Food Chemistry*, **2018,** *245*(Supplement C), 426-431.

[134] Chikhoune, A.; Gagaoua, M.; Nanema, K.D.; Souleymane, A.S.; Hafid, K.; Aliane, K.; Hadjal, S.; Madani, K.; Sentandreu, E.; Sentandreu, M.Á.; Boudjellal, A.; Križman, M.; Vovk, I. Antioxidant Activity of Hibiscus sabdariffa Extracts Incorporated in an Emulsion System Containing Whey Proteins: Oxidative Stability and Polyphenol–Whey Proteins Interactions. *Arabian Journal for Science and Engineering*, **2017,** *42*(6), 2247-2260.