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Natural pigments from plant sources: A review

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Abstract

Pigments are present in all living matter and provide attractive colors and play basic but important roles in the development of organisms. Human beings, like most animals, come in contact with their surroundings through color and things can or cannot be acceptable based on their color characteristics. The names of many common pigments reflect historical discoveries rather than a set naming system. The carotene was first isolated from *Daucus carota* (carrot), violaxanthin from *Viola tricolor* (pansy), and the common anthocyanidins, pelargonidin, cyanidin, peonidin, delphinidin, petunidin and malvidin from *Pelargonium, Centaurea, Paeonia, Delphinium, Petunia* and *Malva*, respectively. In this paper a review of the various plants pigments have been made. The pigments are in leaves, fruits, vegetables, and flowers and also present in skin, eyes and other animal structures and in bacteria and fungi. Natural and synthetic pigments are used in medicines, foods, clothes, furniture, cosmetics, and in other products. The most noticeable and widespread pigments of plants are chlorophylls. These are cyclic tetrapyrrole pigments chelated with magnesium, and they share structural features with the haem and bile pigments of animals. The flavonoids are phenylpropanoid compounds of widespread occurrence. There are several major classes of flavonoids; however, only a few of these provide pigments to plants, in particular the anthocyanins and proanthocyanidins (condensed tannins).

Keywords: Plant pigments, carotenoids, terpenoid, anthocyanins, betalains, flavonoid, Natural food colourants, Betalains

Introduction

Pigments are present in all living matter and provide attractive colors and play basic but important roles in the development of organisms. Human beings, like most animals, come in contact with their surroundings through color and things can or cannot be acceptable based on their color characteristics. Many biological structures, such as skin, eyes, fur and hair contain pigments such as melanin in specialized cells called chromatophores.

Humans without colour blindness can detect wavelengths between approximately 380 and 730 nm, representing the visible spectrum of red, orange, yellow, green, blue, indigo and violet. So, chlorophyll with maximum absorbencies at 430 and 680 nm will leave wavelengths forming a green colour. Often the colours are the result of a mix of residual wavelengths; for example, anthocyaninsm absorbing yellow-green light wavelengths of 520–530 nm will generate mauve colours formed by the reflection of a mix of orange, red and blue wavelengths. Thus the pigments can be described in two ways: the wavelength of maximum absorbance (lmax) and the colour perceived by humans (Davies, 2004) [24].

Plant pigmentation is generated by the electronic structure of the pigment interacting with sunlight to alter the wavelengths that are either transmitted or reflected by the plant tissue. The specific colour perceived will depend on the abilities of the observer. In recent decades, there has been growing interest in the identification and characterization of high added value extracts from natural sources that are capable of providing additional benefits to human health. Such compounds include antioxidants, anti-inflammatory compounds and antihypertensive agents, among others.

The names of many common pigments reflect historical discoveries rather than a set naming system. For example, carotene was first isolated from *Daucus carota* (carrot), violaxanthin from *Viola tricolor* (pansy), and the common anthocyanidins, pelargonidin, cyanidin, peonidin, delphinidin, petunidin and malvidin from *Pelargonium, Centaurea, Paeonia, Delphinium, Petunia* and *Malva*, respectively. More complete names, giving details of constitution and stereochemistry, have been developed for many compounds to meet the standards of the International Union of Pure and Applied Chemistry (IUPAC) and International Union of Biochemistry (IUB) (Delgado-Vargas *et al.*, 2000) [10].

The pigments are in leaves, fruits, vegetables, and flowers and also present in skin, eyes and other animal structures and in bacteria and fungi. Natural and synthetic pigments are used in medicines, foods, clothes, furniture, cosmetics, and in other products. However, natural pigments have important functions other than the imparted beauty, such as the following: we could not have photosynthesis or probably life all over the world without chlorophylls and carotenoids. Under stress conditions plants show the synthesis of flavonoids; the quinones are very important in the conversion of light into chemical energy.

Classification

1. **By Their Origin:** Pigments can be classified by their origin as natural, synthetic, or inorganic. Natural pigments are produced by living organisms such as plants, animals, fungi, and microorganisms. Synthetic pigments are obtained from laboratories. Natural and synthetic pigments are organic compounds. Inorganic pigments can be found in nature or reproduced by synthesis (Delgado-Vargas and Paredes-Lopez 2003) [9].
2. **By the Chemical Structure of the Chromophore:** Pigments can be classified by taking into account the chromophore chemical structure as: Chromophores with conjugated systems: carotenoids, anthocyanins, betalains, caramel, synthetic pigments, and lakes. Metal-coordinated porphyrins: myoglobin, chlorophyll and their derivatives (Krishna, 2008) [22].
3. **By the Structural Characteristics of the Natural Pigments:** The natural pigments can be classified by their structural characteristics as: (Delgado-Vargas and Paredes-Lopez 2003) [9]. Tetrapyrrole derivatives: chlorophylls and heme colors. Isoprenoid derivatives: carotenoids and iridoids. *N*-heterocyclic compounds different from tetrapyrroles: purines, pterins, flavins, phenazines, phenoxazines, and betalains. Benzopyran derivatives (oxygenated heterocyclic compounds): anthocyanins and other flavonoid pigments. Quinones: benzoquinone, naphthoquinone, anthraquinone, Melanins.
4. **As Food Additives:** By considering the pigments as food additives, their classification by the FDA is
   a. **Certifiable:** These are manmade and subdivided as synthetic pigments and lakes.
   b. **Exempt from certification:** This group includes pigments derived from natural sources such as vegetables, minerals, or animals, and manmade counterparts of natural derivatives.

Plant pigments

Plant pigments exist in many varied forms, some with highly complex and large structures. For example, over 600 naturally occurring carotenoid structures have been identified (Britton, 1995) [6] and over 7000 flavonoids, including over 500 anthocyanins. The complexity of some pigments is well illustrated by the anthocyanin Ternatin A1, which consists of the base 15-carbon anthocyanin modified with seven molecules of glucose, four molecules of 4-coumaric acid and one molecule of malonic acid, corresponding to $C_{96}H_{107}O^{+}_{53}$ (Delgado-Vargas and Paredes-Lopez 2003) [9]. The plant pigments are grouped on a common structural and biosynthetic basis into four major groups (Table 1). In addition to these major groups there is a great array of pigments that are of limited taxonomic occurrence, and often poorly characterized.

The most noticeable and widespread pigments of plants are chlorophylls. These are cyclic tetrapyrrole pigments chelated with magnesium, and they share structural features with the haem and bile pigments of animals. The carotenoids also associated with photosynthesis and additionally providing bright colours to flowers and fruits. Carotenoids are terpenoid pigments present in all photosynthetic plants and they also occur in photosynthetic bacteria such as Erwinia and Rhodobacter. Annual production of carotenoids by plants, algae and dinoflagellates has been estimated at 100 million tons (Britton, 1995) [6]. The flavonoids are phenylpropanoid compounds of widespread occurrence. There are several major classes of flavonoids; however, only a few of these provide pigments to plants, in particular the anthocyanins and proanthocyanidins (condensed tannins). The betalains are nitrogenous pigments that are the most taxonomically restricted of the major plant pigment groups, being found only in a few families of the order Caryophyllales and some fungi. Their occurrence is mutually exclusive to that of the anthocyanins.

Within plants, the major pigment groups show wide occurrence in the different tissues. For example, flavonoids occur in almost all tissues, carotenoids in leaves, roots, tubers, seeds, fruits and flowers, and even chlorophylls occur in flowers and fruits as well as leaves. Within tissues, there is often distinct localisation of pigment types in different cell layers. Anthocyanins are typically found in epidermal cells in petals and sub-epidermally in leaves, and chlorophyll in the sub-epidermal photosynthetic cell layers of leaves. The sub-cellular localisation of the different pigment groups is also generally distinct. The chlorophylls and carotenoids are principally lipid-soluble, plastid-located pigments, although there are examples of water-soluble carotenoids, at least some of which are located in the vacuole via plastid–vacuole interactions (Bouvier *et al.*, 2003a) [3]. The betalains are water-soluble and vacuolar-located while flavonoids occur in many sub-cellular locations, as well as extracellularly, the coloured flavonoids are principally found in the vacuole (Bohm, 1998) [2]. For flavonoids, the sub-cellular localisation is just one of several factors that determine the behaviour of the pigment molecule in the cell and the colour generated from it (Brouillard and Dangles, 1993) [7]. The mechanisms by which pigments such as the flavonoids are directed to the correct subcellular compartment are poorly defined, although some of the steps for anthocyanins have been elucidated (Winefield, 2002) [35]. Interactions of plant pigments with other cellular compounds have been well defined for flavonoids and small molecules (Brouillard and Dangles, 1993) [7], and it is known that both flavonoids and carotenoids interact with specific proteins in the cell (Vishnevetsky *et al.*, 1999, Winefield, 2002) [34, 35].

Functions of pigments in plants

1) In vegetative tissues

Chlorophylls and carotenoids are very important in photosynthesis, chlorophylls capture the light energy and as the primary electron donors and carotenoids works as an essential structural components of the photosynthetic apparatus, where they protect against photo-oxidation. Plant pigments are also involved in other interactions of plants with light, in particular the response to UV radiation.

Anthocyanins also frequently occur in vegetative tissues. The most spectacular example is their contribution to autumn colours in leaves of many deciduous species, which they generate in combination with the retention of carotenoids and loss of chlorophyll (Matile, 2000; Hoch *et al*., 2001; Lee, 2002) [29, 21, 27]. In non-senescing tissues their occurrence is more sporadic. Some species accumulate them in significant amounts in healthy leaves, providing red or purple colours to the foliage. In other cases anthocyanin production is induced in leaves in response to stresses such as cold, high light levels, pest and pathogen attack or deficiency of nutrients such as phosphate and nitrogen. Anthocyanin colouration in leaves can vary with season, environment, between individuals of a population and between different leaves on a single plant. It is commonly thought that anthocyanins have a role in protecting the photosynthetic apparatus from damage in many of these situations and those tissues that show more anthocyanin accumulation are often at greater photo-inhibitory risk, e.g. during nutrient reabsorption in senescing leaves or in cold temperatures (Hoch *et al*., 2001) [21]. But, the details of how anthocyanins achieve this are not determined. One hypothesis is that anthocyanins help attenuate the light levels, modifying the quantity and quality of light incident on the chloroplasts and thus reducing excitation pressure.

Anthocyanins are acting as both direct light screens under high light stress and general antioxidants against harmful reactive oxygen species in the various other stress situations in which they are prevalent (Delgado-Vargas and Paredes-Lopez 2003) [9]. It was also observed that the red-leafed morphs of some shade species have a significant antioxidant advantage over green morphs, that anthocyanins can enhance oxidative protection in species more directly exposed to the sun and that anthocyanins can reduce photoinhibition and photobleaching of chlorophyll under light stress conditions.

The role of anthocyanins in improving foliar nutrient re-absorption during senescence, through the shielding of the photosynthetic apparatus from excess light, was tested using wild-type and anthocyanin-deficient mutants of three deciduous woody species under varying environmental conditions (Hoch *et al*., 2003) [20]. Nitrogen re-absorption efficiencies of the mutants were significantly lower than the wild-type counterparts, supporting the protection hypothesis of anthocyanins in senescing leaves. Anthocynin has a role in protecting light-sensitive phototoxic plant defense compounds from degradation.

2. In Reproductive tissues

The most observable function of plant pigments is to provide colour to flowers and fruit for attraction of pollinators and seed dispersal agents. These colours arise predominantly from flavonoid and carotenoid pigments and a short guide to the likely pigments producing specific colours in flowers and fruits of plants (Table 2). There are also few common pigments that generate colours in specific species. For angiosperms, colour is key to attracting pollinators, whether they are bees, butterflies, other insects or birds. Even if, it is frequently one of a number of factors, including fragrance, floral shape which combine to determine pollinator choice. Flavonoids are the most common flower colour pigments. The role of flavonoids in pollination was the subject of an extensive review by Harborne and Grayer (1994) [17]. They noted the shortage of detailed studies on specific pigments and pollinators. This has changed greatly in the last decade, with many studies determining pollinator preference with regard to individual colours, fragrances and even petal epidermal cell shape.

For pollinators such as bees that can detect light in the UV spectrum, UV absorbing pigments also influence flower selection. The main contributors to the UV absorbance of the flower are the chalcone- and flavonol-type flavonoids.

Flavonols are very common in flowers, often being in greater abundance than the coloured pigments. The flavonoids may form UV-visible patterning in petals, often in combination with UV-reflective carotenoid pigments (Harborne & Grayer, 1994; Bohm, 1998) [17, 2].

Carotenoids and flavonoids are commonly colour pollen. They have been shown to have a role in signaling to pollinators (Lunau, 2000) [28], and it is possible they also have protective activities against various stresses. Colourless flavonoids are known to be involved in plant fertility in some species (Taylor and Jorgensen, 1992; Jorgensen *et al*., 2002) [32, 23], but this has not been shown for coloured flavonoids. The role of pigments in fruit is in signaling the ripeness of the fruit to seed-dispersal agents. Both carotenoids and flavonoids commonly provide fruit colours.

Flavonoids and carotenoids can also colour seeds, e.g. the yellow carotenoids and purple flavonoids of maize kernels.

Economic aspects of plant pigments

The most noticeable application of pigments is being for tattooing (henna) and carthamin, indigo and other pigments to generate bright colours in clothing. The use of carthamin extract (from safflower, Carthamus tinctorius) to dye the wrappings of mummies has been reported (Gilbert & Cooke, 2001) [14]. It was also reported that the tanning of animal leathers with polymeric phenols was used in garment manufacturing. Now days, there are tanneries carrying out leather treatment and dyeing with plant pigments.

From an economic perspective, putting to one side the vital role of chlorophylls and carotenoids in photosynthesis, the most obvious contribution of plant pigments to agriculture is with regard to consumer choice of fresh fruit, vegetables and floriculture products. They are also of economic importance as flavour and colour components of teas, wine and other beverages, as natural food colourants, for the health of ruminant animals, as plant defense agents and for amelioration of damaging UV light. The great range of non-coloured flavonoids and alkaloids produced by plants is often key to plant defence, and the biosynthesis of these compounds, and their importance to plant defence, agriculture and the pharmaceutical industries, Wink, 1999a, 1999b [36, 37]

Natural food colourants

The quality of food is firstly assessed by its visual characteristics such as colour. Fresh food is highly coloured by the major plant pigment groups, like carotenoids and anthocyanins in fruit and chlorophylls in green vegetables. However, the pigmentation is often lost during manufacturing of processed foods and the visual appeal of the final product is enhanced using added colourants. Before the discovery of synthetic dyes, the food industry was solely reliant on natural food colourants. The use of natural colourants in many applications was superseded by synthetic dyes, in recent years industries has been returned to use the natural colourants with increased interest in new and improved sources in food applications.

Plant pigment widely used as food colourants are: annatto, anthocyanins, betalains (beetroot pigment) and curcumin (turmeric pigment). Together with the insect-derived pigment cochineal, they account for over 90% of the market for natural food colourants (Hendry, 1996) [19]. Use of chlorophyll as a food colourant is very limited in comparison to these pigments, principally because of its poor stability during food processing or in response to light or acid conditions in the final food product.

Important Plant Pigments

1. Anthocyanins

Anthocyanins are a group of plant pigments that are widely distributed in nature. They generally occur in the plant as glycosides and acylglycosides of anthocyanidins, the aglycones. Anthocyanidins vary in the different hydroxyl or methoxyl substitutions in their basic flavylium (2-phenylbenzopyrilium) structure. Anthocyanins have been demonstrated to play a very important role in plant physiology and are important to the food industry and in human health. (Wu and Prior, 2005) [39] Thus, understanding the chemical structures of anthocyanins and their distribution in foods are critical to anthocyanin-related studies. These are the most abundant, visible to the human eye and widespread of the flavonoid pigments. They absorb light at the longest wavelengths and are the basis for most orange, pink, red, magenta, purple, blue and blue-black floral colours. Key to providing such colour diversity is the degree of oxygenation of the anthocyanidins (central chromophores of anthocyanins) and the nature and number of substituent (e.g. sugar moieties) added to these chromophores. At a primary level, the degree of oxygenation has the greatest impact on the colour of anthocyanin pigments. Most anthocyanins are derived from just three basic anthocyanidin types: pelargonidin, cyanidin and delphinidin.

Anthocyanins are responsible for many of the attractive colors, from scarlet to blue, of flowers, fruits, leaves, and storage organs. They are almost universal in higher plants, but in general anthocyanins seem absent in the liverworts, algae, and other lower plants, although some of them have been identified in mosses and ferns. The type of anthocyanins in plants is so variable that some ornamental plants present only one main type of anthocyanin *(Dianthus, Petunia*), whereas others have mixtures *(Rosa, Tulipa,* and *Verbena*). On the other hand, some fruits are a source of one anthocyanin: cyanidin in apple, cherry, fig, and peach; delphinidin in eggplant and pomegranate; some fruits have two main anthocyanins such as cherry sweet and cranberry (cyanidin and peonidin), while others have several anthocyanins (grape). In general, the anthocyanin concentration in most of the fruits and vegetables goes from 0.1 up to 1% d.w.

Anthocyanins are vacuolar pigments and in this organelle the presence of membrane bound bodies called anthocyanoplasts has been proposed; these structures are formed while pigment synthesis is in operation, and eventually they are dispersed to produce a totally pigmented vacuole. In flowers, anthocyanins are almost exclusively located in epidermal cells, and only occasionally in the mesophyll. In case of the leaves of rye *(Secale cereale*) they are restricted to the mesophyll cells. The copigmentation mechanism is unique to the anthocyanin family. Anthocyanins also react with alkaloids, amino acids, benzoic acids, coumarin, cinnamic acids, and a wide variety of other flavylium compounds. This weak association is termed intermolecular co-pigmentation. Intra-molecular co-pigmentation is due to the acylation in the molecule, and it is more effective than intermolecular; in acylated anthocyanins, it is suggested that acyl groups interact with the basic anthocyanin structure, avoiding the formation of the hydrated species. The basic role of copigments is to protect he colored flavylium cation from the nucleophilic attack of the water molecule (Baublis and Berber-Jiménez, 1995, Saito *et al.* 1995) [1].

Functions

a) **Colour and Ecological Functions:** As flavonoids, anthocyanins are benzopyran derivatives. Thus, anthocyanins showed functions in plants as antioxidant, photoprotection, defense mechanism, as well as other ecological functions (symbiosis phenomena). In particular, anthocyanins are the most important pigmenting compounds between the flavonoids, and consequently they show an interesting role in several reproductive mechanisms of plants such as pollination, seed dispersal, and antifeedant. Additionally, anthocyanins have been proposed as taxonomic.

b) **Practices in Food Processing:** Anthocyanins have been used to evaluate the adulteration of some pigmented food products (Boyles and Wrolstad, 1993) [4]. Prune juice is a product in which brown color is developed by the reaction of phenolic compounds and anthocyanins, and it is possible the adulteration of prune juice with other fruit juices improve its color. To control this possible source of adulteration, it is believed that prune juice can have only traces of anthocyanins, while the adulterated juice will show increased levels (van Gorsel, 1992) [33]. Also, anthocyanin is used to determine the authenticity of fruit jams. With this kind of analyses, it was determined that labeled black cherry jams in reality were prepared with common red cherries (less expensive fruit). In addition, it was suggested that adulteration of blackberry jams with strawberries can be detected with analysis of the relation between pelargonidin and cyanidin 3-glucoside. It was also noted that this methodology is very efficient because anthocyanins are pretty stable during jam manufacture.

c) **Pharmacological Effects:** It is believed that humans are well conditioned to large consumptions of these compounds. Anthocyanin daily intake was in the range 25 to 215 mg/person, depending on gender and age, and this intake is largely enough to induce pharmacological effects. The consumption of wine flavonoids has been correlated with low incidences of coronary hearth diseases (French paradox) and similarly chokeberry (*Anonia melanocarpa*) extracts have shown very strong nutraceutical properties. Moreover, anthocyanins possess bactericidal, antiviral, and fungistatic activities. They exhibit a strong antioxidant activity that prevents the oxidation of ascorbic acid, provides protection against free radicals, shows inhibitory activity against oxidative enzymes, and has been considered as important agents in reducing the risk of cancer and heart disease (Bridle and Timberlake, 1997) [5]. In particular, bioflavonoids have shown activities to improve the permeability and strength of capillaries, to accelerate the ethanol metabolism, and to reduce inflammatory and edematic reactions (Lozovik, G). Similar effects have been observed with crude extracts of *Rubus occidentalis, Sambucus nigra,* and *Vaccinium myrtillus* (Harborne and Grayer, 1988 and Francis, 1989) [16, 12].

Importance as Food Colours

a) **Stability in Model Systems:** Anthocyanins are of great economic importance as fruit pigments and thus also as pigments of fruit juices and wines (Harborne and Grayer, 1988) [16]. Interestingly, one of the main claims of the food industry is for natural colorants to replace synthetic red dyes, and anthocyanins are the principal candidates, but enocyanin and lees (sediment of the grape juice tanks) preparations are the only anthocyanin sources approved by FDA (US) to be used for human food, while the main use is in the production of beverages and soft drinks (Bridle and Timberlake, 1997, Francis, 1989) [5, 12]. However, anthocyanin instability limits their use and different preparations have been evaluated to avoid anthocyanin degradation. The color of anthocyanins is provided by its resonating structure, but resonance phenomena also confer its intrinsic instability. Moreover, it has been established that instability has a direct relation with the number of hydroxyl groups and indirect with the number of methoxyl groups.

The equilibrium colored-uncolored anthocyanin structures affect the stability of products on storage, because the R^+ form is the most stable.

$$R^+ + H_2O \longrightarrow ROH + H$$

Colored Catonic Form Uncolored Pseudobase Form

In solution, anthocyanin molecules are present in equilibrium between the colored cationic form and the colorless pseudobase. This equilibrium is directly influenced by pH. Acidic pH is favorable for the colored form that diminished with pH increments. Some anthocyanins are red in acid solutions, violet or purple in neutral solutions, and blue in alkaline pH. This is the reason that most colorants containing anthocyanins can only be used at pH values below four (Delgado-Vargas *et al.*, 2000) [10].

b) **Processing and Stability in Foods:** Anthocyanin pigments can be destroyed easily during the processing of fruits and vegetables, and considering food color as an appealing characteristic, many studies have been carried out to understand the anthocyanin properties and to obtain better products with minimal degradation. High temperature, increased sugar level, pH, and ascorbic acid can affect the rate of destruction. Temperature has been reported to induce a logarithmic destruction of pigment with time of heating at a constant temperature. Bleaching by effect of heat occurs because of the above-described equilibrium is changed toward the uncolored forms. It has been suggested that flavonoid structure is opened to form chalcone, which is degraded further to brown products. However, it has been observed that optimal conditions permit the regaining of color on cooling if there is sufficient time (several hours) for the reconversion.

2. Carotenoids

Carotenoid pigments provide distinctive red, orange and yellow colours and a number of carotenoid-derived aromas to many fruits and flowers, making them commercially important in agriculture, food manufacturing and the cosmetic industry. However, it is their roles in photosynthesis and nutrition that account for the absolute requirement for carotenoids in the survival of plants and mammals alike. Specifically, carotenoids are a ubiquitous component of all photosynthetic organisms as they are required for assembly and function of the photosynthetic apparatus. Carotenoids are also a vital part of our diet as antioxidants and precursors to vit A.

Structure

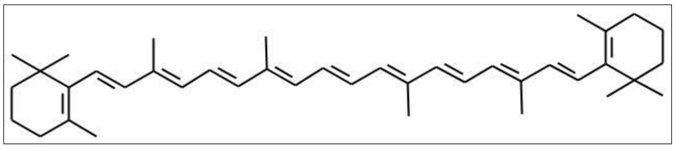
Carotenoids are isoprenoid compounds, biosynthesized by tail to tail linkage of two C_{20} geranylgeranyl diphosphate molecules. This produces the parent C_{40} carbon skeleton from which all individual variations are derived. This skeleton can be modified by: Cyclization at one end or both ends of the molecule to give different end groups, Changes in hydrogenation levels and addition of oxygen containing functional groups. Carotenoids that contain one or more oxygen atoms are known as xanthophylls, the parent hydrocarbon as carotene. The long system of alternating double and single bonds constitutes a conjugated system in which the p electrons are effectively delocalised over the entire length of the polyene chain (Figure 1). This feature is responsible for the molecular shape, chemical reactivity and light absorbing properties, and hence color of carotenoids (Britton, 1995, Dutta *et al.*, 2005) [13, 11].

Functions

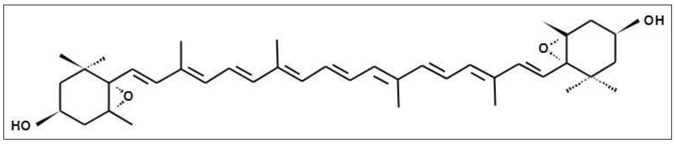
a. **Colour:** Carotenoids provide colors to flowers, seeds, fruits and to some fungi and colour has an important role in reproduction: coloration attracts animals that disperse pollen, seeds, or spores. In *Phycomyces blakesleanus* it was observed that intracellular accumulation of excess Carotenoids disturb the mating recognition system, which appears to be involved in the later stages of mating by inhibiting the cell-to-cell recognition systems (Delgado-Vargas *et al.*, 2000) [10].

b. **Photosynthesis:** Main pigments involved in photosynthesis are chlorophylls and carotenoids. Carotenoids have two well-known functions in photosynthesis: (1) accessory pigments in light harvesting and (2) as photoprotectors against oxidative damages. It has been proposed that carotenoids as light harvesting compounds evolved from anaerobic organisms, then generalized to all of the aerobic photosynthetic organisms. One of the carotenoid structural characteristics is their ability to absorb visible light: p delocalized electrons suffer a photo-induced transformation in which a singlet state (S_2) is produced, and then energy is efficiently transferred to chlorophyll (chl) to form singlet chl with a slightly higher energy. Carotenoid functions are greatly determined by their associated proteins. These proteins are mainly membranal, usually hydrophobic, which bound carotenoids by noncovalent bonds (Gottfried *et al.*, 1991) [15].

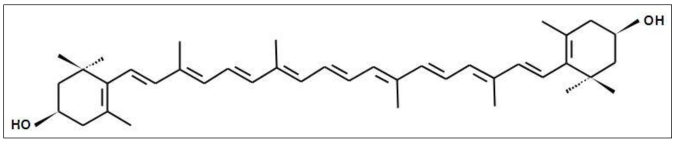
c. **Antioxidant:** The research studies have shown that carotenoid photoprotective role is related to its antioxidant activity or with modulation of other cellular antioxidants. Also, it has been established that carotenoid structure has a great influence in its antioxidant activity; for example, canthaxanthin and astaxanthin show better antioxidant activitythan β-carotene or zeaxanthin. Antioxidant activity of capsanthin and lutein was evaluated using chlorophyll as photosensitizer. Capsanthin was a better antioxidant, and it was concluded that the antioxidant activity depended on the number of double bonds, keto groups, and that cyclopentane rings in the carotenoid structure enhanced their activity. It was suggested that carotenoids can be used in foods to prevent degradation of other components (Chen *et al.*, 1994) [8].



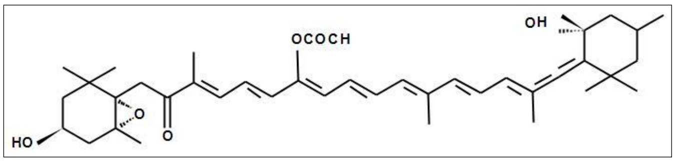
β-carotene



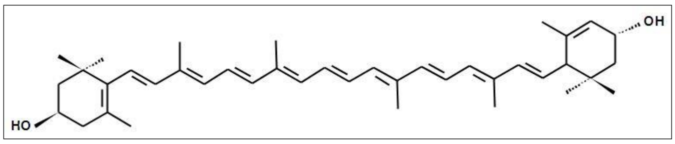
Violaxanthin



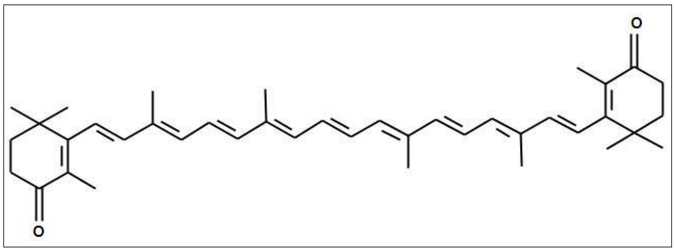
Zeaxanthin



Fucoxanthin



Lutein



Canthaxanthin

Fig 1: Structure of common Carotenoids (Delgado-Vargas *et al.*, 2000).

Importance as Food Colors

Carotenoids have been used as food colors for centuries: saffron, pepper, leaves, and red palm oil have carotenoids as their main color components. Color of carotenoids, together with beneficial properties such as vitamin A precursors and antioxidants; have led to their wide application in the food industry; preparations to apply them in oily or aqueous media have been produced, including emulsions, colloidal suspensions, and complexes with proteins. These preparations have found applications to pigment margarine, butter, fruit juices and beverages, canned soups, dairy and related products, deserts and mixes, preserves and syrups, sugar and flour confectionery, salad dressings, meat, pasta and egg products, among others and other important areas of application of carotenoids have been emerged.

a) Processing and Stability in Foods

Processing effects were analyzed in tomato and green vegetables (broccoli, spinach, and green beans). Moderate processing did not produce Carotenoid modifications, but a prolonged heating (1 h boiling) conduces to total destruction of epoxycarotenoids; it was shown that under this condition only the most sensitive carotenoids are highly destroyed. Boiled tomato showed an identical carotenoid profile with fresh tomato, and differences in content were only detected. It was observed that drying process produces a net carotenoid biosynthesis that is enhanced by illumination: drying under darkness conditions increased the carotenoid content in 15%, while under illumination it was 47%. The biosynthesis occurred in a first stage (35 to 40% of moisture), then degradation was observed. The effect of mango processing on Carotenoid content was evaluated. Mango was sliced and stored at vacuum or frozen (-40°C) conditions during 6 months; other samples were commercially canned. Fresh and frozen fruits showed similar characteristics. Canned mango showed great changes in color and carotenoid profile. The most stable carotenoid was β-carotene. Chen *et al*., stated that the effect of juice carrot processing on its carotenoid content. Pasteurization (105°C/25 s) neither produced a considerable variation in the isomeric profile nor in the Carotenoid content.
Blanching (98°C /5 min), cooking (98°C for 15, 30, and 60 min) and sun drying (25°C ± 6°C) are traditional processing practices. In general, blanching resulted in a reduction of β-carotene and in an increment of α-carotene concentration. Cooking enhanced the quantity of carotenoids extracted, and sun drying reduced the concentration of carotenoids in all evaluated vegetables. Also, it was shown that thermal processing increased the vitamin A activity of all vegetables, but in amaranth leaves a reduction was observed. Thus, blanching and cooking could be considered as advantageous processes for increasing the amount of pro-vitamin A available when vegetables are consumed.

For the preservation of color in carotenoid pigmented foods, the antidiscoloring effect of green tea polyphenols in beverages and in margarine was assayed. In general, it was observed that tea polyphenols and several tea catechins have stronger effects against the discoloration of β-carotene than L-ascorbate (widely used to prevent discoloration) in the aqueous (beverages) and oily (margarine) systems.

Marigold pigments have also been used in the pigmentation of shrimp *(Penaeus vannamei*). It was found that saponified extracts were better pigmenting agents than esterified marigold extracts. Its study suggested that lutein and zeaxanthin (main carotenoids of marigold) can be metabolized into astaxanthin and deposited in shrimp and better colorations could be obtained than with astaxanthin supplemented feed. In the pigmentation of whitefleshed fish, carotenoids must be eliminated from the corn gluten meal (CGM) used in feeds.

3. Betalains

Betalains are the yellow and violet pigments that substitute anthocyanins in plants belonging to the order Caryophyllales. These pigments have attracted much attention because of their bioactivities, which range from an antioxidant capacity to the chemoprevention of cancer. They are water-soluble, but unlike anthocyanins they are indole-derived compounds synthesized from tyrosine. This class of pigments is found only in the Caryophyllales (including *cactus* and *amaranth*), and never co-occur in plants with anthocyanins. Betalains are responsible for the deep red colour of beets, and are used commercially as food-coloring agents.

Functions

a) Taxonomic Markers

Considering the crucial dependence of a living system on the economy of energy and our growing knowledge about the chemoecology of secondary products in nature, this assumption can be feasible.

b) Ecological and Physiological Aspects

As in the case of other secondary metabolites, it is impossible to assign a definite function to betalains in the economy of the organisms that produce them. When pigments are in flowers or fruits they may have a role as attractants for vectors (insects or birds) in the pollination process and in seed dispersal by animals, such as anthocyanins. The occurrence in other plant parts (e.g., leaves, stem, and root) may be devoid of immediate function. However, it has been suggested that betalain accumulation in red beet root is related to the storage of carbohydrates as a physiological response under stress conditions (Kolb, 1997) [25]. In addition, the transient coloration of many seedlings and the reddening of senescent leaves of several plants of Caryophyllales order (e.g., *Kochia scoparia*) have no obvious physiological or ecological reasons. Whatever its significance, the process resembles the analogous phenomenon observed in anthocyanin- producing species.

Betalains are also produced in injured tissues, normally not pigmented, possibly as a defense mechanism against infection. This physiological response was only observed in plants possessing specific factors that have been associated with two novel antifugal proteins (Kragh *et al., 1995)* [26].

Importance as Food Colors

a) Stability in Model Systems

When betalains are used as food colorants, color stability is a major concern. There are several factors that have been recognized to affect the stability of these pigments, pH. Betalain solutions in this pH range showed a similar visible for betacyanins and betaxanthins.

b) Processing and Stability in Foods

The sensitivity of betalains to different factors suggests that their application as food colorants is limited. Based on these properties, betalains can be used in foods with a short shelf-life, produced by a minimum heat treatment, and packaged and marketed in a dry state under reduced levels of light, oxygen, and humidity. Betalains have several applications in foods, such as gelatins desserts, confectioneries, dry mixes, poultry, dairy, and meat products. The amount of pure pigment required in these foods groups to obtain the desired hue is relatively small and for most applications does not exceed 50 ppm of betalains, calculated as betanin. Problems associated with betalain degradation and pigment recovery during the processing operations are of economic importance and must be solved to betalains displace the application of synthetic dyes in some food products. The effectiveness of commercial betalains depends largely on a continuous availability of highly pigmented sources, the use of cold and modified storage atmospheres prior to processing, efficient enzymatic control, handling practices, extraction procedures, purification, concentration, and finishing operations e.g., freeze, spray, and vacuum drying).

Table 1: Major pigment of plants and their occurrence in other organisms

S. No.	Pigments	Common Type	Occurrence
1	Betalains	Betacyanins Betaxanthins	The Caryophyllales and some fungi
2	Carotenoids	Carotenes Xanthophylls	Photosynthetic plants and bacteria Retained from the diet by some birds, fish, and crustaceans
3	Chlorophylls	Chlorophyll	All photosynthetic plants
4	Flavonoids	Anthocyanins Aurones Chalcones Flavonols Proanthocyanidins	Widespread and common in plants, including angiosperms, gymnosperms, ferns, fern allies and Bryophytes. Retained from the diet by some insects

(Davies, 2004)

Table 2: Most common pigment types associated with flower and fruit colours in plants

Colour	Pigment type	Pigment group	Examples
Cream	Flavonols or flavones	Flavonoid	Cream flowers
Pink to red		Carotenoid	Some red flowers and fruit, e.g. *Lycopersicon esculentum* (tomato) fruit
	Pelargonidin and/or cyanidin	Flavonoid	Most pink flowers and some fruit, e.g. *Eustoma grandiflorum* (lisianthus) flowers
	Pelargonidin and/or cyanidin	Flavonoid	Most red flowers and some fruit, e.g. *Malus* (apple) fruit
	Anthocyanin and carotenoid mix	Flavonoid and carotenoid	e.g. *Tulipa* flowers
	Betacyanin	Betalain	In the Caryophyllales, e.g. *Bougainvillea* flowers
Orange		Carotenoid	Most orange flowers and fruit, e.g. *Tagetes erecta* (marigold) flowers
	Pelargonidin Alone	Flavonoid	A few examples, e.g. *Pelargonium* flowers
	Anthocyanin and aurone mix	Flavonoid	Rare occurrence, e.g. *Antirrhinum majus* (snapdragon) flowers
	Anthocyanin and chalcone mix	Flavonoid	Rare occurrence, e.g. *Dianthus* (carnation) flowers
	Betacyanin	Betalain	A few examples in the Caryophyllales, e.g. *Portulaca* (purslane) flowers
Yellow		Carotenoid	Most yellow flowers and fruit
	Aurone	Flavonoid	Rare occurrence, e.g. *Antirrhinum majus* flowers
	Chalcone	Flavonoid	Rare occurrence, e.g. *Dianthus* flowers
	Flavonol	Flavonoid	Rare occurrence, e.g. *Gossypium* (cotton) flowers
	Betaxanthin	Betalain	A few examples in the Caryophyllales, e.g. *Portulaca* flowers
Green		Chlorophyll	All green flowers and fruit
Blue	Delphinidin	Flavonoid	Most blue flowers and fruit

	Cyanidin	Flavonoid	Rare occurrence, e.g. *Ipomoea* (morning glory) flowers
Purple		Carotenoid	Rare occurrence, e.g. *Capsicum* (pepper) fruit
	Cyanidin and/or delphinidin	Flavonoid Flavonoid and carotenoid mix	Most mauve flowers, e.g. *Petunia* and some purple fruit, e.g. *Solanum melongena*, Some flowers, e.g. *Cymbidium* orchids
Black	Delphinidin	Flavonoid and carotenoid mix	Some black flowers, e.g. *Viola* (pansy)

(Davies, 2004).

Table 3: Plant species frequently utilized as source of dying materials

S. No.	Name of plant	Part (organ) of the plant	Main colour	Pigment
1	Nettle	Young Leaf	Green	Chlorophyll
2	Mayweed	Flower	Yellow	Flavonoid glycosides
3	Annatto	Seed	Yellow, Orange	Carotinoids
4	Safflower	Petal	Yellow, Orange	Calconderivate carthamin, Safflor-yellow
5	Smoke tree	Shoot, Leaf	Yellow	Tannins, Flavonoids
6	Saffron	Stigma	Yellow, Orange	Carotinoids
7	Turmeric	Root	Yellow	Curcumin (1,7hepadiene-3,5dion)
8	Greenweed	Shoot	Yellow	Isoflavonoid genistein
9	St. John's wort	Flowering shoot	Yellow Red	Hypericine and derivatives
10	Weld	Flowering shoot	Yellow	Flavonoid glycosides
11	Saw wort	Flowering shoot	Yellow	Flavonoid glycosides
12	Golden rod	Flower	Yellow	Flavonoid glycosides
13	African marigold	Flower	Yellow	Xantophyll
14	Rainfarn	Flowering shoot	Yellow	Flavonoid glycosides
15	Mullein	Petal, leaf	Yellow	Flavonoid glycosides
16	Alkanet	Root	Red	Naphtoquinones
17	Hollyhock	Petal	Red, Violet	Anthocyanin
18	Beetroot	Root	Red, Violet	Betalaines (peptidderivatives)
19	Oregano	Flowering shoot	Red, Brown	Tannins, Flavonoide
20	Red sounder	Wood	Red	Santalins (benzoxanthemon derivative)
21	Madder	Root	Red	Anthraquinone (alizarin)
22	Grape	Berry peel	Red, Violet	Anthocyanin
23	Logwood	Wood	Blue, Black	Haematoxilin
24	Indigo	Leaf, Stem	Blue	Peptide-glycoside (precursor of indigo)
25	Woad	Leaf	Blue	Peptide-glycoside (precursor of indigo)
26	Elder	Berries	Violet, Blue	Anthocyanin (sambucyanin)
27	Agrimony	Flowering shoot	Brown	Catechines, Flavonoides
28	Onion	Bulb, Leaves	Rusty-Brown	Carotinoids
29	Walnut	Green fruit shell	Brown, Black	Naphtoquinones
30	Common alder	Bark	Black	Naphtoquinones

(Nemeth, Encyclopedia of Life Support Systems (EOLSS) http://www.eolss.net/Eolss-sampleAllChapter.aspx in: Cultivated plants, primarily as food sources-Vol. II-Colouring (Dye) Plants-Eva Nemeth)

Conclusions

Plant pigmentation is generated by the electronic structure of the pigment interacting with sunlight to alter the wavelengths that are either transmitted or reflected by the plant tissue. The specific colour perceived will depend on the abilities of the observer. The pigments are in leaves, fruits, vegetables, and flowers and also present in skin, eyes and other animal structures and in bacteria and fungi. Natural and synthetic pigments are used in medicines, foods, clothes, furniture, cosmetics, and in other products. The complexity of some pigments is well illustrated by the anthocyanin Ternatin A1, which consists of the base 15-carbon anthocyanin modified with seven molecules of glucose, four molecules of 4-coumaric acid and one molecule of malonic acid, corresponding to $C_{96}H_{107}O^{+}_{53}$. Carotenoids are terpenoid pigments present in all photosynthetic plants and they also occur in photosynthetic bacteria such as Erwinia and Rhodobacter. Anthocyanins also frequently occur in vegetative tissues. The most spectacular example is their contribution to autumn colours in leaves of many deciduous species, which they generate in combination with the retention of carotenoids and loss of chlorophyll. The role of anthocyanins in improving foliar nutrient re-absorption during senescence, through the shielding of the photosynthetic apparatus from excess light, was tested using wild-type and anthocyanin-deficient mutants of three deciduous woody species under varying environmental conditions. The quality of food is firstly assessed by its visual characteristics such as colour. Fresh food is highly coloured by the major plant pigment groups, like carotenoids and anthocyanins in fruit and chlorophylls in green vegetables. However, the pigmentation is often lost during manufacturing of processed foods and the visual appeal of the final product is enhanced using added colourants.

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