Chapter 9
Pigments

Kewei Chen

Abstract This chapter gives an overall introduction of food pigments including their roles in the food industry, coloring mechanisms behind their chemical nature and their classification. Based on the chemical structures, some important and common natural pigments are illustrated according to their chemical molecules, changes and treatments during food processing, and related protective techniques including heme, chlorophylls, carotenoids and flavonoids with anthocyanins, catechins and tannins emphasized. In this chapter, plenty of detailed information about surrounding environmental effects on the changes of pigment molecules and the related color is provided to help readers understand the reaction mechanisms and to associate them with food processing techniques such as acid/alkaline treatment, modified atmospheric package and heat treatment.

Keywords Pigment · Food color · Heme · Chlorophyll · Carotenoid · Flavonoid · Anthocyanin · Catechin · Tannin

9.1 Overview

9.1.1 Definition of Pigments and Their Role in the Food Industry

The color of a substance is due to its capacity of selectively absorbing some visible light, thus reflecting the rest of the unabsorbed visible light that visualizes in human eyes. So, the substances in food that show this kind of characteristics leading to the presence of colorful food are collectively referred to as food pigments, including natural pigments inherent in food materials, colored substances formed in food processing, and added food colorants. Food colorants are natural or synthetic chemicals that have been subjected to stringent safety assessment tests and approved for use in the food coloring process.

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Table 9.1 Food color and associated sensory evaluation for consumers

Color	Sensory evaluation	Color	Sensory evaluation
Red	Strong flavor, ripe, delicious	Gray	Unpalatable, dirty
Yellow	Fragrant, bland, ripe, delicious	Purple	Strong, sweet, warm
Orange	Sweet, nourishing, strong, delicious	Light Brown	Unpalatable, hard, warm
Green	Fresh, refreshing, cool, acid	Dark Orange	Old, hard, warm
Blue	Fresh, refreshing, cool, acid	Cream	Sweet, nourishing, refreshing, delicious
Coffee	Unique flavor, rich texture	Dark Yellow	Not fresh, unpalatable
White	Nutritious, refreshing, hygienic, soft	Light Yellow-Green	Refreshing, cool
Pink	Sweet, soft	Yellow-Green	Refreshing, cool

Color is one of the main sensory indicators for food. Before acquirement of other food information, consumers often judge the food by the color to have a primary picture of food quality, freshness or maturity. For example, the color of a fruit is related to maturity, and the color of fresh meat is inseparable from its freshness. Therefore, how to improve the color characteristics of food is very crucial, which should be taken into consideration by food producers and processors. Food color that meets consumers' psychological requirements can give consumers beautiful enjoyment and increase their appetite and desire to buy.

The color of food can stimulate sensory organs of consumers and make them associate with the taste (Table 9.1). For example, red gives a feeling of ripeness and good taste that people generally like, and therefore plenty of candy, pastries and beverages are in red.

Besides, color can affect people's perception of food flavor. For example, red beverages are considered to have the flavors of strawberry, black strawberry, or cherry; yellow beverages are related to lemon flavor, and green beverages are referred to as lime flavor. Therefore, in the beverage industry, different flavors of beverages are often given different colors that meet the psychological requirements of consumers.

What's more, brightly colored foods can increase appetite. The color from red to orange is most appetizing, and light green and turquoise can also increase appetite, while yellow green depresses, as it is related with the long-term cognition for food, e.g., red apples, orange tangerines, yellow cakes and verdant vegetables. The color of some spoiled foods can make people feel bored, so some colors that are not too bright generally give a bad impression. Even if the same color is used in different foods, it will produce different feelings. For example, purple grape juice is widely accepted, but no one likes purple milk.

The color of foods is mainly decided by their inherent pigments. For example, the color of meat is mainly determined by myoglobin and its derivatives; the color of green leafy vegetables is mainly composed of chlorophyll and its derivatives. In food storage, the color change is often encountered, which sometimes is desirable. For instance, the color becomes more attractive during fruit ripening, and the bread turns out to be a brownish yellow color during the baking process. However, more often it needs to be avoided. For example, the browning on the cut surface after the apple is sliced; green vegetables become brownish green after cooking, and raw meat loses fresh red and gets brown during storage. Most of these changes in food color are due to chemical changes in food pigments. Therefore, understanding different food pigments is of great significance for controlling food color.

In food processing, the control of food color is usually carried out by two methods: color preservation and dyeing. Color protection means less loss of pigment, which calls for raw materials with appropriate maturity, proper processing technology, no excess acidic, alkaline or heat treatment, no access to metal equipment, less exposure to oxygen, etc. Dyeing is another common method to obtain and maintain the desired color for food. Since a coloring agent can produce plenty of color combinations, and its stability is better than inherent pigment, it is very convenient to apply it in food processing. However, the use of synthetic dyeing additives always arouses safety concerns, and it is necessary to comply with food safety regulations and food additive standards to prevent the abuse of colorants.

9.1.2 Mechanism of Food Coloration

Different substances can absorb light with different wavelengths. If the wavelength of light absorbed by a substance is outside the visible light region, the substance appears colorless; if in the visible light region (400–800 nm), the substance will exhibit a certain color. Color is related to the wavelength of the reflected light that is not absorbed. The color seen by the human eye is a composite color composed of visible light with different wavelengths reflected by the object. For example, if an object absorbs only invisible light and reflects all visible light, then it appears colorless; conversely, it appears black or nearly black. When the object selectively absorbs part of visible light, its color is represented by a composite color composed of unabsorbed visible light (also known as the complementary color of the absorbed light wave). Figure 9.1 gives a comparison of different light wavelengths, related colors and complementary colors.

Food pigments are generally organic compounds that often have ***chromophores***, which have an absorption peak in the ultraviolet and visible light regions (200–800 nm), in their molecular structure. Always, a chromophore contains a conjugation system with multiple –C=C– bonds, and/or –C=O, –N=N–, –N=O, –C=S combinations. When a molecule contains a chromophore with an absorption wavelength from 200 to 400 nm, the substance is colorless, whereas if a molecule contains two or more
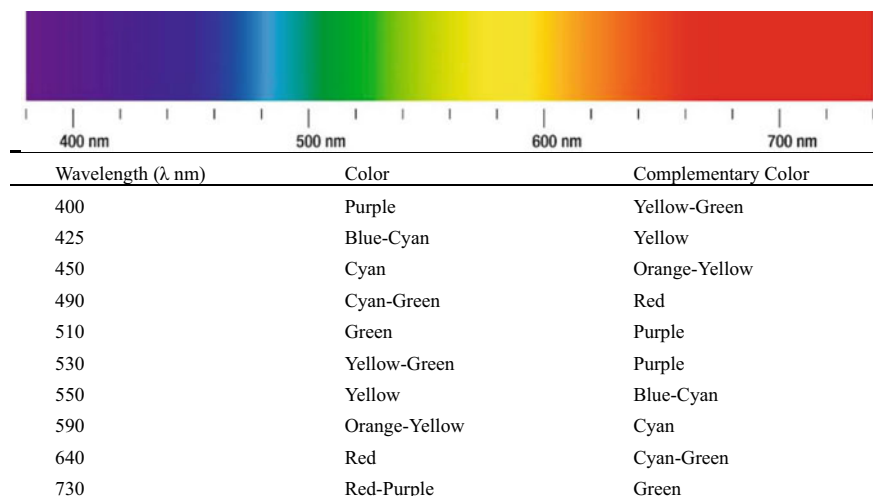
Wavelength (λ nm)	Color	Complementary Color
400	Purple	Yellow-Green
425	Blue-Cyan	Yellow
450	Cyan	Orange-Yellow
490	Cyan-Green	Red
510	Green	Purple
530	Yellow-Green	Purple
550	Yellow	Blue-Cyan
590	Orange-Yellow	Cyan
640	Red	Cyan-Green
730	Red-Purple	Green

Fig. 9.1 Light wavelengths, related colors and complementary color

chromophores, the absorbed light moves from a short wavelength to a long wavelength, causing the substance to develop color. The larger the conjugated system, the longer the wavelength absorbed by the structure, as can be seen by the example given in Table 9.2.

Otherwise, there are also some chemical groups, such as –OH, –OR, –NH$_2$, –SR$_2$, –SR, –CI and –Br, whose absorption bands are in the ultraviolet region and they do not produce color by themselves, but when combined with conjugated systems or attached with chromophores, the absorption wavelength of the entire molecule can be shifted to the long-wave direction, thus producing a color. This kind of group is called ***auxochrome*** and it helps to modify the color presentation of a compound. For example, according to a different arrangement of groups such as –OH and –OCH$_3$ on 2-phenylbenzofuran parent ring, with modified positions and

Table 9.2 Relationship between the maximum absorption wavelength (λ) by conjugated polyene compounds and the number of double bonds

Compounds	Conjugated double bonds (number)	Absorption wavelength (nm)	Color
Butadiene	2	217	Colorless
Hexatriene	3	258	Colorless
Dimethyloctene	4	296	Light Yellow
Vitamin A	5	335	Light Yellow
Dihydro- β-Carotene	8	415	Orange
Lycopene	11	470	Red
Dehydrolycopene	15	504	Purple

group numbers, various anthocyanins form. The structure of food colorants contains chromophores and auxochromes. Understanding their structure and properties is of great significance to the research, development and use of colorants.

9.1.3 Classification of Food Pigments

Food pigments can be divided into natural pigments and synthetic dyes. Natural pigments contain plant pigments such as chlorophyll, carotenoids and anthocyanins, animal pigments such as heme, carotenoids in yolk and shrimp shells, and microbial pigments such as monascus pigments.

Natural pigments can be classified into azoles (or porphyrins), isoprenes, polyphenols, ketones and quinones depending on their chemical nature. For instance, porphyrin pigments include chlorophyll and heme and isoprene pigments contain carotenoids. Anthocyanins and flavonoids belong to polyphenols and ketones, respectively; and shellac colors and cochineal are quinone pigments.

Synthetic pigments can be classified into azo-based pigments and non-azo-based pigments depending on whether the molecule contains a –N=N-chromophore structure. For example, carmine and tartrazine are azo pigments, while etythrosine and brilliant blue are non-azo pigments.

Further, food pigments can be classified into water-soluble and liposoluble decided by their solubility properties. The former are always synthetic pigments for the application in the food industry, while the latter are always natural pigments. In this chapter, natural pigments will be introduced according to their structures and changes during food processing and storage.

9.2 Tetrapyrrole Pigments

Tetrapyrrole pigments are characteristic of their planar macrocycle where four cyclic or linear pyrrole groups are connected. In the food industry, tetrapyrrole pigments are represented by chlorophyll and heme, with the former found widely in green fruits, vegetables and algae and the latter in charge of the color changes of meat and related products.

9.2.1 Chlorophyll

9.2.1.1 Structures and Properties

Chlorophyll is the main pigment in a photosynthetic unit such as green plants, algae and photosynthetic bacteria that helps to capture light for photosynthetic reactions.
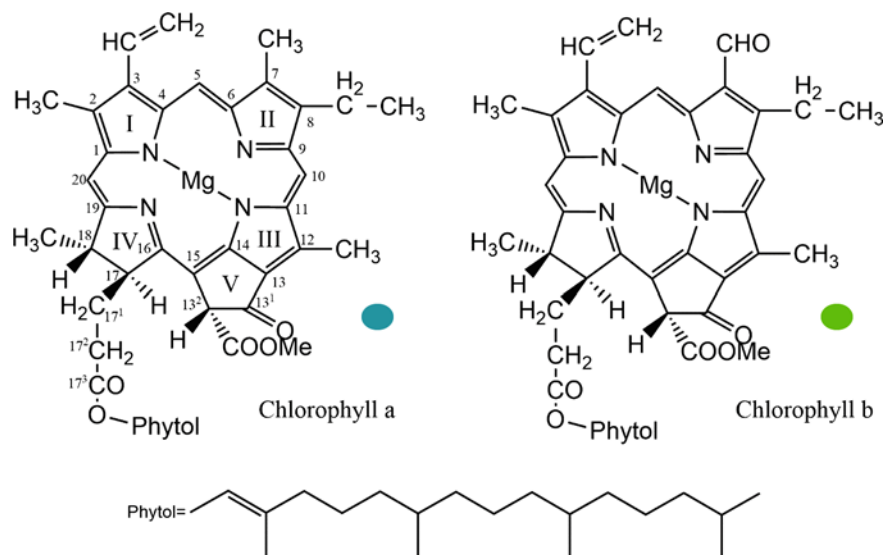
Fig. 9.2 Chemical structures of chlorophyll a and chlorophyll b

Higher plants, only contain chlorophyll a and chlorophyll b, and additionally, marine algae have chlorophyll c and chlorophyll d. Photosynthetic bacteria contain bacteria chlorophyll. This chapter mainly introduces chlorophyll a and chlorophyll b present in higher plants, and their structures are shown in Fig. 9.2.

Chlorophyll is a magnesium-containing tetrapyrrole derivative, which is linked by a four-pyrrole ring and four methylal groups (–CH=) into a macrocycle, also called porphyrin. The magnesium atom is in the center of the porphyrin ring that is easily substituted by two hydrogen atoms and tends to be positively charged, while the surrounding nitrogen atoms tend to have a negative concentration, so porphyrins are polar and can bind to proteins. Additionally, an isocyclic ring V is formed on the porphyrin ring which is quite unstable and involves many reactions. A phytol chain is esterified with propionic acid moiety at C_{17}, and this phytol chain is also called the "tail" of chlorophyll, which is a diterpene composed of 4 isoprene units so that the phytol chain decides the lipophilic nature of chlorophyll compounds. The structural difference between chlorophyll a and chlorophyll b is only that the substituents at the C_3 position are different: chlorophyll a contains a methyl group, chlorophyll b contains a formyl group (Fig. 9.2), and the ratio between chlorophyll a and chlorophyll b in higher plants is around 3 to 1.

Pure chlorophyll a is a black-green powder that is soluble in ethanol solution with a blue-green solution and has deep red fluorescence. Chlorophyll b is a green powder whose ethanol solution is yellowish-green and shows fluorescence. Both are insoluble in water and soluble in organic solvents, and chlorophyll pigments are often extracted from plant homogenates by organic solvents such as acetone, ethanol and ethyl acetate. Chlorophyll in plants is generally present in the chloroplasts, which are

complexed with carotenoids, lipids and lipoproteins and distributed on disk-shaped lamellar membranes.

In food processing and storage, ***chlorophyll*** pigments are chemically transformed into several important derivatives, including phytol-free derivatives (***chlorophyllide***), magnesium-free derivatives (***pheophytin*** and ***pheophorbide***), pyro-chlorophyll derivatives (***pyropheophytin***, and ***pyropheophorbide***), and oxidized derivatives (***C13^2-hydroxy chlorophyll*** derivatives and ***C15^1-hydroxy lactone chlorophyll*** derivatives). Some derivatives have been modified with several reactions and Fig. 9.3 illustrates the main reactions that happened to chlorophyll pigments during food processing.

Take chlorophyll a, for instance. The phytol ester bond can be hydrolyzed by the action of chlorophyllase to form phytol and chlorophyllide a that is hydrophilic. Chlorophyll a and chlorophyllide a transform to pheophytin a and pheophorbide a, respectively, when magnesium ions are detached under heating or acidic conditions. In addition, the isocyclic ring V in the chlorophyll molecule is associated with various chemical reactions under heating and aerobic conditions including the following: (1) the methyl ester group at the C13^2 position is susceptible to conformational changes in isolated solutions, forming chlorophyll epimer; (2) the methyl ester group is removed under heating to form pyrochlorophyll derivatives; and (3) oxidation reaction. The hydrogen atom at position C13^2 is easily oxidized to the hydroxy group under aerobic conditions or with oxidase to form C13^2-hydroxy chlorophyll derivatives; besides, at the same time, ring V forms a lactone structure to form C15^1-hydroxyl lactone chlorophyll derivatives. In some cases, ring V forms a cyclic anhydride structure, that is, purpurin-18 a derivative. If the cyclic tetrapyrrole structure of chlorophyll a is opened, the chlorophyll structure becomes linear and yields colorless chlorophyll catabolites.

The various reactions that occur to chlorophyll pigments mean changes in molecular structure and related color. Taking chlorophyll acetone solution as an example, chlorophyll a is blue-green, and if magnesium is detached from the tetrapyrrole ring, the color changes to brown immediately. The pure loss of phytol chain or methyl ester group and oxidation of C13^2-H to C13^2-OH does not affect the chlorophyll molecular tetrapyrrole structure or chlorophyll color. The spectral absorption peak of C15^1-hydroxy lactone chlorophyll a is shifted by 10 nm to the short-wave direction, and the color slightly changes, while the purpurin-18 a chlorophyll derivative is purple in color. In the processing of green vegetables and fruits, chlorophyll reactions are always complexed with many factors, resulting in a de-green phenomenon for food manufacturing.

9.2.1.2 Changes in Chlorophyll in Food Processing and Storage

1. Enzymes

There are two types of enzymatic changes that cause chlorophyll degradation. The enzyme that directly uses chlorophyll as a substrate is only chlorophyllase, which is
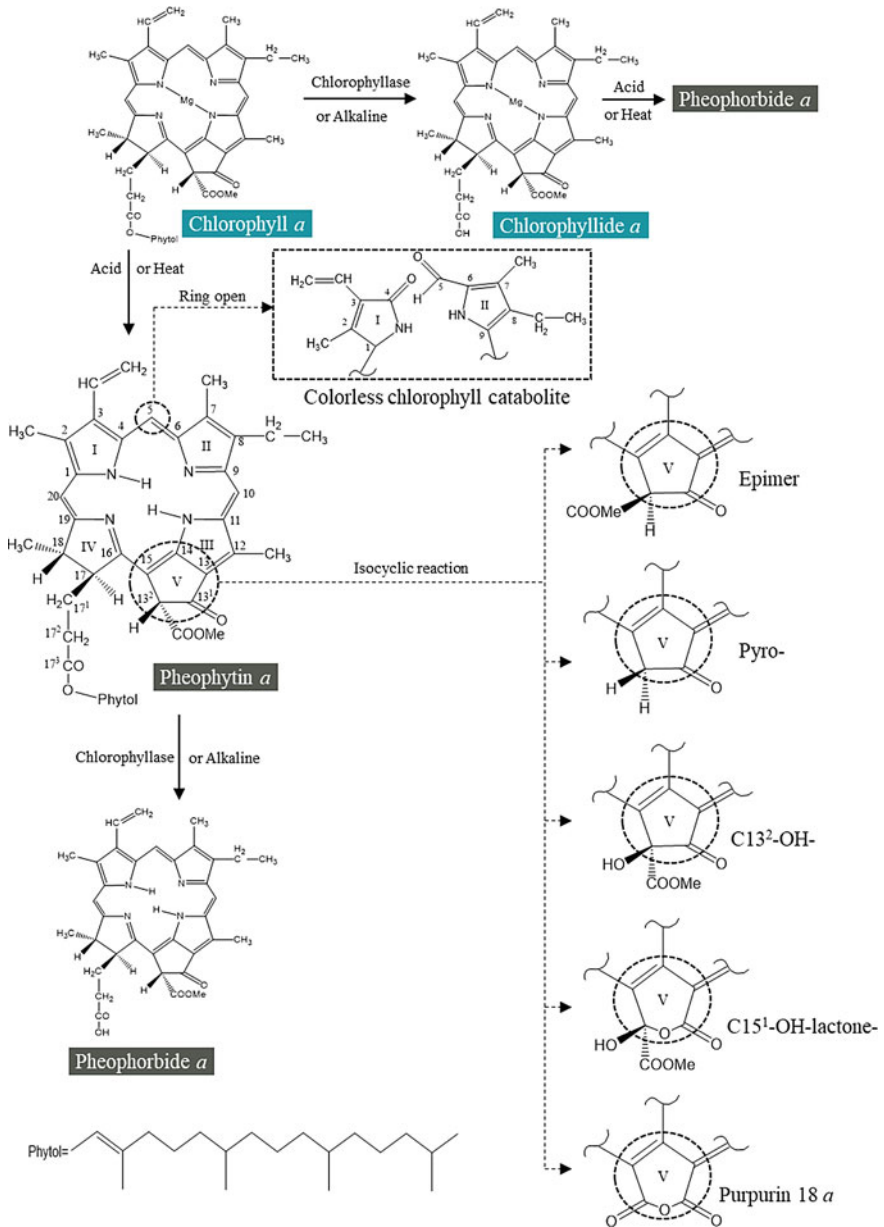
Fig. 9.3 Chlorophyll derivatives and their chemical changes during processing conditions

an esterase that catalyzes the hydrolysis of phytate ester bonds of chlorophyll and pheophytin to produce chlorophyllide and pheophorbide, respectively. The optimum temperature of chlorophyllase is in the range of 60–80 °C, and its activity begins to decrease above 80 °C and can be fully depressed by 100 °C. Indirect enzymes include lipase, protease, pectinesterase, lipoxygenase, peroxidase, etc. Lipase and protease act to destroy the chlorophyll-lipoprotein complex, so that chlorophyll pigments are more susceptible to reactions. The role of pectinesterase is to hydrolyze pectin to pectic acid, thereby lowering the pH of the system and transforming chlorophyll into pheophytin. Lipoxygenase and peroxidase can catalyze their respective substrates and the yielding intermediates cause oxidative degradation of chlorophyll pigments.

2. Heat and acid

The green color of green vegetables after cooking or blanching seems to be strengthened and it may be due to the gas existing in the intercellular space that is heated out, or that the distribution of different components in the chloroplast changes during the thermal process. But during heat treatment, protein denaturation in the chlorophyll-lipoprotein complex causes chlorophyll to be separated. Free chlorophyll is very unstable and sensitive to light, heat and enzymes. At the same time, the tissue cells are destroyed during the heating process, resulting in increased permeability of hydrogen ions across the cell membrane, hydrolysis of triglyceride into fatty acids and carbon dioxide produced by decarboxylation, all of which induce a pH decrease in the surrounding environment.

pH is an important factor in determining the rate of magnesium removal in the chlorophyll molecule. Chlorophyll is relatively stable to heat at pH 9, and easily magnesium-dechelated at pH 3. The decrease in pH induces the formation of pheophytin in plant cells and further produces pyropheophytin, causing the green color of the food to change significantly to brown, and this transformation is irreversible in an aqueous solution. Chlorophyll a is faster in this reaction than chlorophyll b, as the positive charge in the porphyrin ring of chlorophyll b is relatively more, which increases the difficulty of magnesium detachment and makes it more stable than chlorophyll a.

Treating tobacco leaves with NaCl, $MgCl_2$ or $CaCl_2$ and heating them to 90 °C could slow down the magnesium removal reaction by 47%, 70% and 77%, respectively. The role of salt may be as an electrostatic shielding agent. Cations neutralize the negative charge of fatty acids and proteins on the chloroplast membrane, thereby reducing the rate of protons permeating the membrane. The use of cationic surfactants also has a similar effect. It adsorbs to the chloroplast or cell membrane, and restricts the diffusion of protons into the chloroplast, thereby slowing down the effect of magnesium removal.

The previous view believed that the thermal stability of chlorophyllide was higher than that of chlorophyll, but later studies proved the opposite. It is now believed that phytol has a steric hindrance on the substitution of protons for magnesium atom, so chlorophyllide is easier to have magnesium detachment than chlorophyll. In addition, chlorophyllide is water-soluble and will be more likely to encounter protons to cause the magnesium reaction. When enzyme, acid and heat conditions are combined, the
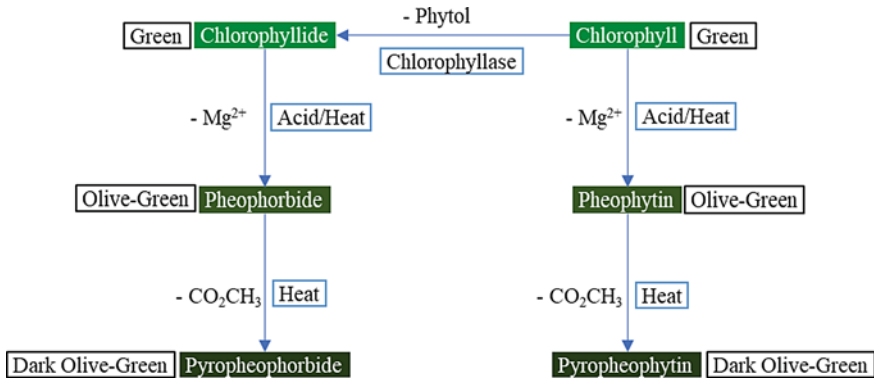
Fig. 9.4 Chlorophyll reactions under enzyme, acid and heat conditions

sequence of chlorophyll changes which can be seen in Fig. 9.4. These reactions are quite common in the curing process of green vegetables, as large sums of lactic acid are accumulated during fermentation.

3. Light

In fresh plants, chlorophyll and protein are combined in the form of protein complexes and they participate in photosynthesis reaction without photodegradation; but when plants are senescent during storage and processing, pigments are exposed to photodegradation due to the cell membrane damage. Under aerobic conditions, chlorophyll or porphyrins can produce singlet oxygen and hydroxyl radicals when exposed to light, which can react with chlorophyll tetrapyrroles to form peroxides and more radicals, and ultimately lead to the decomposition of the porphyrin ring and complete loss of color. The photolytic process of chlorophyll pigments begins with the ring-opening of the methylene group and the main product is glycerin, along with a small amount of lactic acid, citric acid, succinic acid and malonic acid, as shown in Fig. 9.5.
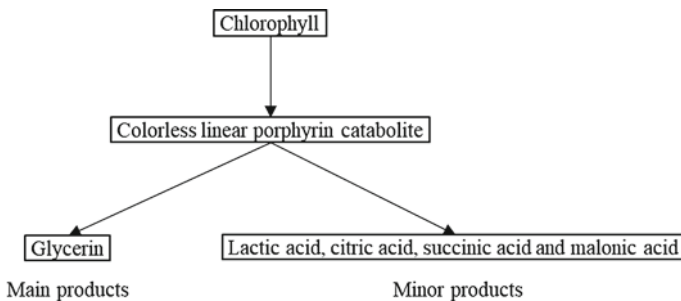

Fig. 9.5 Chlorophyll reactions under light condition

9.2.1.3   Protective Measurements of Chlorophyll Pigments

1. Acid control

Increasing the pH of canned vegetables is an effective method for color preservation. The application of calcium oxide and sodium dihydrogen phosphate to maintain the pH of the blanching solution close to 7.0, or the use of magnesium carbonate, sodium carbonate and sodium phosphate, is an effective method for chlorophyll preservation. However, they are partly limited due to the softening of vegetable tissues and produce an alkali odor.

The traditional Blair method is to use calcium hydroxide or magnesium hydroxide in the blanching solution to increase the pH and maintain the brittleness of vegetables, while this method can only delay chlorophyll loss up to around 2 months as the inside of canned vegetables cannot be treated effectively. Alternatively, coating the inner wall of the tank with ethyl cellulose containing 5% magnesium hydroxide can slowly release the magnesium hydroxide into the food to maintain the pH value of 8.0 for a long time so that the green color can be kept for a relatively long time. A disadvantage of this method is that it will cause partial hydrolysis of glutamine and asparagines to produce an ammonia odor, and cause hydrolysis of lipids. In green peas, this preservative method may also cause the formation of struvite.

2. HTST

High-temperature short-time sterilization (HTST) not only preserves vitamins and flavors, but also significantly reduces the green damage in commercial sterilization. However, after about 2 months of storage, the chlorophyll pigments will also degrade due to the pH decline. It is more effective when HTST is combined with pH adjustment; however, the color protection effect that has been achieved will also fade away due to the drop in pH during storage.

3. Green reoccurrence

Adding zinc ions to the bleaching solution of vegetables is also an effective method for protecting green. The principle is that pheophytin can chelate zinc ions to form zinc metallo complexes (mainly zinc pheophytin and zinc pyropheophytin). This method can produce satisfactory results for the processing of canned vegetables. This method uses a zinc concentration of about a few ten thousandths, and the pH value is controlled at about 6.0, and the heat treatment is performed at a temperature slightly higher than 60 °C. To improve the permeability of zinc in the cell membrane, an appropriate amount of surface-active anionic compounds can also be added to the treatment solution. This method can produce satisfactory results when used in the processing of canned vegetables. Zinc ions can be substituted by copper ions.

Another chlorophyll additive in the food industry is called copper chlorophyllin or zinc chlorophyllin which is widely allowed in Asia and European countries for the improvement of processed vegetable color.

4. Others

The modified atmosphere preservation technology enables the green color to be protected along with the preservation of fresh vegetables. When the water activity is very low, even if acid is present, the chance of H^+ transfer and access to chlorophyll is relatively reduced, so it is not easy to replace Mg^{2+} of chlorophyll pigments. At the same time, due to low water activity, microbial growth and enzyme activity are also inhibited. Therefore, dehydrated vegetables can remain green in color for a long period. When storing green plant foods, separation from light and oxygen can prevent oxidative fading of chlorophylls. Therefore, the proper option of packaging materials and chlorophyll preservative methods, combined with the appropriate use of antioxidants, can maintain the green color of green vegetables and fruits for a long time.

9.2.2   Heme

9.2.2.1   Structures and Properties

Hemes are the main red pigments in animal muscles and blood, mainly present in the form of myoglobin in the muscle and hemoglobin in the blood. The protein part is called globin which consists of 153 amino acid residues. Fe^{2+} in the center of the porphyrin ring has six coordination sites, four of which are coordinated with the nitrogen atom from the tetrapyrrole rings. Another one is coordinately bonded with the histidine residue from the globulin, leaving the sixth coordination site that could be coordinated to small molecules such as O_2 and CO. The structure of heme is illustrated in Fig. 9.6.

Myoglobin is a globular protein composed of one molecule of heme and one globin which has a single polypeptide, and its relative molecular mass is around 16,700. The main role of myoglobin is to receive and store oxygen transported by hemoglobin in muscle cells, helpful for tissue metabolism. The hemoglobin molecule is a tetramer with two alpha-peptide chains (141 amino acid residues for each chain) and two beta-peptide chains (146 amino acid residues for each chain), and each peptide chain
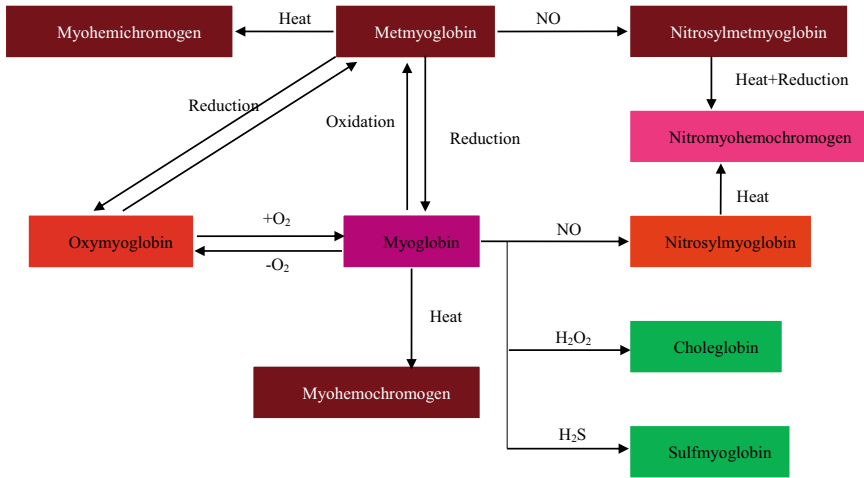
Fig. 9.6   Structure of heme
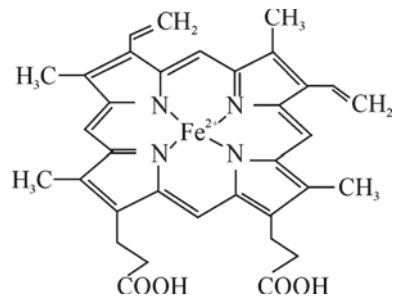
Fig. 9.7 Myoglobin derivatives and related changes during food processing

binds one molecule of heme, with its relative molecular mass around 64,500. The main function is to bind and transport oxygen in the blood.

Nearly 90% of the pigment in the muscle is myoglobin and others are cytochrome enzymes, flavin and vitamin B_{12}. The content of myoglobin in the muscle varies greatly depending on the species, age, sex and location of the animal. For example, the muscle of the veal shows shallow color compared with an adult cow as the lower content of myoglobin. The hemoglobin in shrimps, crabs and insects is hemocyanin containing copper.

In meat processing and storage, myoglobin is converted into a variety of derivatives, the type of which depends mainly on the chemical properties of myoglobin, the valence of iron, the type of ligand of myoglobin and the state of globulin. The heme iron in the porphyrin ring can be in two forms, ferrous iron (Fe^{2+}) and ferric iron (Fe^{3+}). The iron ion of myoglobin is Fe^{2+}, with one coordination site free, and when it combines with oxygen, forms oxymyoglobin. The main derivatives of myoglobin and related reactions are shown in Fig. 9.7 and Table 9.3.

9.2.2.2 Changes in Muscle Color During Storage and Meat Processing

After animal slaughter and bloodletting, due to the stop of oxygen supply to the muscle tissue by hemoglobin, the ***myoglobin*** in the fresh meat maintains its reduced state, and the color of the muscle is purple-red (the color of myoglobin). When the carcass is divided, the myoglobin in the reduced state changes in two different ways as the muscles are in contact with the air. A part of myoglobin reacts with oxygen to form bright-red ***oxymyoglobin***, with typical fresh flesh color; at the same time, another part of myoglobin oxidizes with oxygen to form a brown ***metmyoglobin***,

Table 9.3 Main pigments found in fresh, cured and cooked meat

Compounds	Formation pathway	Iron	Heme ring	Globulin	Color
1. Myoglobin	Reduction of metmyoglobin or deoxygenation of oxymyoglobin	Fe^{2+}	Intact	Natural	Purplish red
2. Oxymyoglobin	Oxygenation of myoglobin	Fe^{2+}	Intact	Natural	Bright red
3. Metmyoglobin	Oxidation of myoglobin and oxymyoglobin	Fe^{3+}	Intact	Natural	Brown
4. Nitrosylmyoglobin	Combination of myoglobin to NO	Fe^{2+}	Intact	Natural	Bright red (pink)
5. Nitrosylmetmyoglobin	Combination of metmyoglobin to NO	Fe^{3+}	Intact	Natural	Dark red
6. Metmyoglobin nitrite	Combination of metmyoglobin to excess nitrite	Fe^{3+}	Intact	Natural	Reddish-brown
7. Myohemochromogen	Myoglobin and oxymyoglobin are treated by heat, denaturing reagents or irradiation	Fe^{2+}	Intact (often combined with non-globulin-type denatured proteins)	Denatured (always isolated)	Dark red
8. Myohemichromogen	Myoglobin, oxymyoglobin, metmyoglobin and myohemochromogen are treated with heating and denaturation reagents	Fe^{3+}	Intact (fften combined with non-globulin-type denatured proteins)	Natural (always isolated)	Brown (sometimes gray)
9. Nitrosohemochromogen	Nitrosylmyoglobin are treated with heating and denaturation reagents	Fe^{2+}	Intact, but one double bond has been saturated	Denatured	Bright red (pink)

(continued)

Table 9.3 (continued)

10. Sulfmyoglobin	Myoglobin interacts with H2S and O2	Fe^{3+}	Intact, but one double bond has been saturated	Natural	Green
11. Sulfmetmyoglobin	Oxidation of sulfmyoglobin	Fe^{3+}	Intact, but one double bond has been saturated	Natural	Red
12. Choleglobin	Myoglobin or oxymyoglobin is treated by hydrogen peroxide; oxymyoglobin interacts with ascorbate or other reducing agents	Fe^2 or Fe^{3+}		Natural	Green
13. Nitrohemin	Co-heating of nitrosylmetmyoglobin and excess nitrite	Fe^{3+}	Intact, but the porphyrin ring is open	None	Green
14. Verdohemin	Excessive denaturing reagent	Fe^{3+}	The porphyrin ring is destroyed	None	Green
15. Bilin	High-dose denaturing reagents	None		None	Yellow or colorless

which is gradually dominated as the flesh color turns brownish-red when the meat is placed in the air for an extended period (Fig. 9.8).

The conversion between myoglobin, oxymyoglobin and metmyoglobin is dynamic and strongly influenced by oxygen pressure. Oxygen pressure is favorable for the formation of oxymyoglobin, and low oxygen pressure is beneficial to the formation of metmyoglobin (Fig. 9.9). In fact, the surface of the freshly cut meat is bright red when it meets enough oxygen. In this case, although there is a certain amount of metmyoglobin production on the surface of the meat, the amount is small. With the storage of meat, the production of metmyoglobin is gradually increased, mainly due to two aspects. On the one hand, there is a small number

Metmyoglobin (Brown)   Myoglobin (Purplish red)   Oxymyoglobin (Bright red)
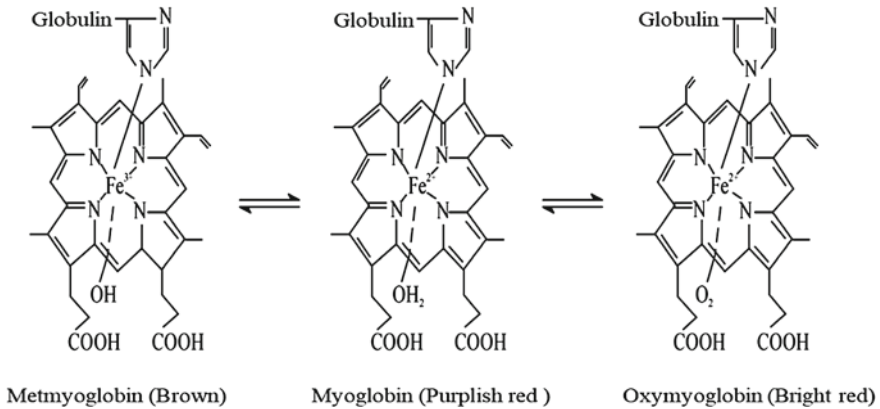
Fig. 9.8 Myoglobin changes in cut meat

Fig. 9.9 The effect of oxygen partial pressure on the conversion among myoglobin, oxymyoglobin and metmyoglobin
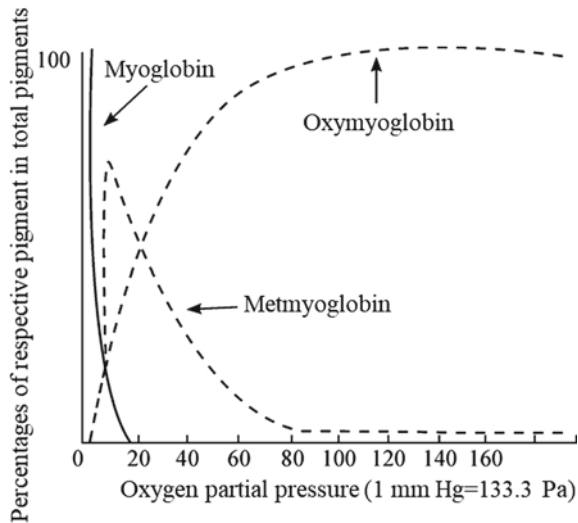
of aerobic microorganisms growing on the surface of the meat, which reduces the oxygen pressure; on the other hand, it is ascribed to the depletion of intrinsic reducing substances such as glutathione and thiol compounds inside the meat which help to reduce metmyoglobin to myoglobin when they are enough.

The reaction from Fe^{2+} in heme to Fe^{3+} is the result of auto-oxidation and the oxidation rate is lower in hemoglobin than in heme, and in oxymyoglobin than in myoglobin, while the auto-oxidation reaction is favored in low pH and with the help of other metal ions such as Cu^{2+}.

When the meat is stored, the myoglobin will be converted into a green substance under certain conditions. This is due to the growth and reproduction of contaminating bacteria that produce hydrogen peroxide or hydrogen sulfide, which react with Fe^{3+} or Fe^{2+} in the heme of myoglobin to produce ***choleglobin*** and ***sulfmyoglobin***, respectively, causing the green color.

$$MbO_2 (Myoglobin) + H_2O_2 \rightarrow Choleglobin \ (Green)$$
$$MbO_2 (Myoglobin) + H_2S + O_2 \rightarrow Sulfmyoglobin \ (Green).$$
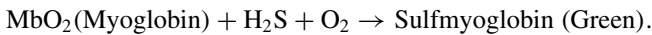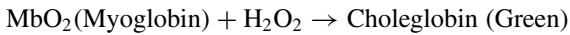
Myoglobin and metmyoglobin are denatured during the heating process. In this case, myoglobin and metmyoglobin yield ***myohemochromogen*** and ***myohemichromogen***, respectively, showing a brown color. That is, during heating, due to the increase of meat temperature and the decrease of oxygen partial pressure, the production of myohemochromogen and myohemichromogen is promoted, and the color of meat changes, especially the production of myohemichromogen, which makes the flesh color turn into brown.

In the processing of meat and salted products such as ham and sausage, nitrate or nitrite is used as a coloring agent. As a result, myoglobin and metmyoglobin are converted into ***nitrosylmyoglobin*** and ***nitrosylmetmyoglobin***, and finally form ***nitromyohemochromogen***, making the color of the cured meat products vivid and attractive, and showing greater tolerance to heat and oxidation. The sixth ligand of the central iron ion of the three pigments is nitric oxide (NO). The reaction pathway is shown in Fig. 9.10.

In addition to the function of a color former, nitrite and nitrate also have the function of preservatives, which is of great significance for the safe storage of meat products. However, the amount of nitrate and nitrite coloring agent must be strictly controlled, because excessive use will not only produce green compounds, but also form carcinogens with certain ammonia species, as shown in Fig. 9.11.

Nitrate and nitrite can form carcinogens with certain ammonia substances. Therefore, in recent years, there have been many studies and reports on the substitutes of nitrate and nitrite, such as the application of monascus colors in fermented sausage, ham, pork luncheon meat and other foods, which can partially replace nitrite. However, currently, no substance has been found that can completely replace nitrite.

Some substances do not have a coloring function, but the use of a coloring agent can significantly improve the color development of nitrates and nitrites, thereby
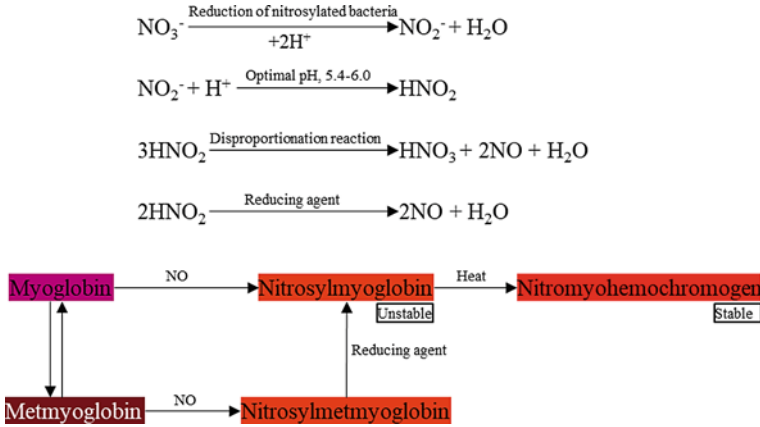
$$NO_3^- \xrightarrow[+2H^+]{\text{Reduction of nitrosylated bacteria}} NO_2^- + H_2O$$

$$NO_2^- + H^+ \xrightarrow{\text{Optimal pH, 5.4-6.0}} HNO_2$$

$$3HNO_2 \xrightarrow{\text{Disproportionation reaction}} HNO_3 + 2NO + H_2O$$

$$2HNO_2 \xrightarrow{\text{Reducing agent}} 2NO + H_2O$$

Fig. 9.10 Muscle color reaction during the curing process

$$RNH_2 + NaNO_2 \xrightarrow{H^+} RNHNO + Na^+ + H_2O$$
$$\boxed{\text{Nitrosamine (carcinogen)}}$$
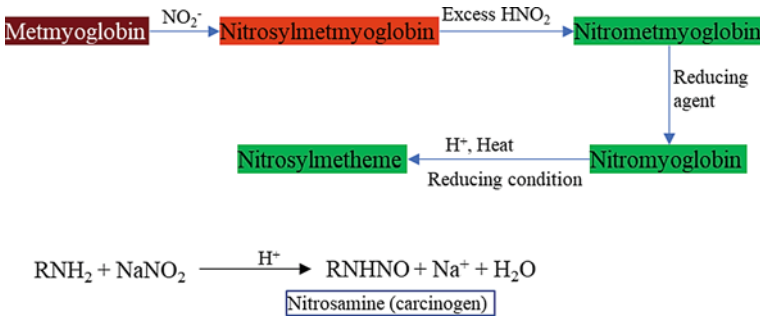
Fig. 9.11 The formation reaction of green substances and carcinogens by excessive use of nitrite

reducing their uses and improving the safety of the meat. Such substances are called coloring auxiliaries that are commonly used in meat products, such as lactic acid, L-ascorbic acid, sodium ascorbic acid and nicotinamide. Lactic acid can promote the formation of nitrous acid; L-ascorbic acid and sodium L-ascorbate can promote the conversion of nitrous acid to nitric oxide. The combination of nicotinamide and myoglobin can form stable nicotinamide myoglobin and prevent myoglobin from getting discolored. By the way, nicotinamide is also a nutrition supplement, with the addition from 0.01 to 0.03%.

Although the color of cured meat products is quite stable under various conditions, visible light can cause them to re-convert to myoglobin and myohemochromogen, while myoglobin and myohemochromogen continue to be oxidized and then converted into metmyoglobin and myohemichromogen. Therefore, bacon products become brown under light. Under the premise of use limit, enough nitrite and antioxidants such as ascorbic acid will help prevent the light browning of cured meat products, as they can re-transform photolytic products into nitrosylmyoglobin and
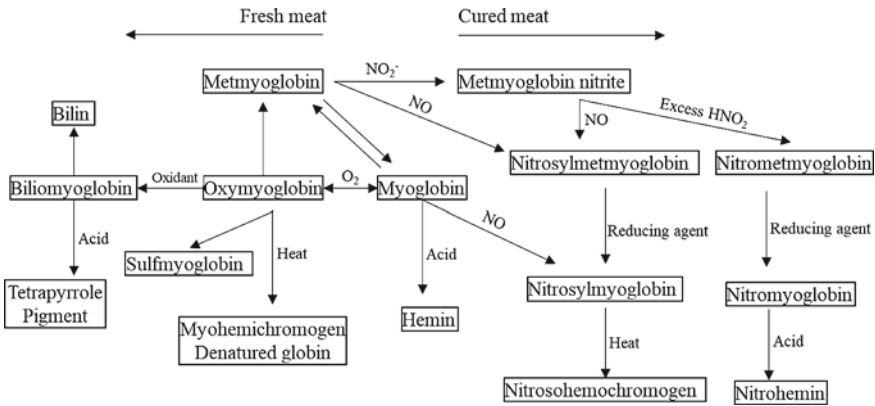
Fig. 9.12 The reaction of myoglobin in fresh meat and cured meat

nitromyohemochromogen. A series of changes in myoglobin in cured meat is shown in Fig. 9.12.

9.2.2.3 Color Protection of Meat and Meat Products

The stability of meat pigments is related to factors such as light, temperature, relative humidity, water activity, pH and microbial growth.

Put the fresh meat in a transparent bag with low air permeability, seal it after vacuuming, and if necessary, add a small amount of oxygen scavenger into the bag to keep the inside oxygen-free. This can make the myoglobin in the meat stay reduced. That is, the iron ions in hemoglobin are Fe^{2+} and have no oxygen combined with them, and the color of the meat can remain unchanged for a long time. Once the bag is opened, a large amount of oxygen is brought into contact with the surface of the meat, and the flesh color is quickly turned to the bright red color of the oxymyoglobin. This is the packaging method for fresh meat in supermarkets.

Modified atmosphere packaging is another effective method for meat or meat products. The color of the meat can be better protected with 100% CO_2 gas. However, when CO_2 pressure is not so high, the meat is prone to brown, mainly due to the conversion of myoglobin to metmyoglobin. If an oxygen scavenger is used in combination, the color protection effect can be improved, but the growth of anaerobic microorganisms must be controlled at the same time.

The color protection method of cured meat products is mainly to protect from light and oxygen. When choosing a packaging method, care must be taken to avoid microbial growth and product water loss. Because choosing the right packaging method can not only ensure the safety of such products and reduce weight loss, but also is one of the important color protection measures.

9.2.2.4 Application of Heme

In the food industry, heme is generally used as a food additive or iron supplement. Compared with iron in plants and other inorganic iron supplements, heme has the advantages of high absorption rate, no toxic side effects, etc., and it has a good clinical effect on iron deficiency anemia.

At present, heme is mainly used in the pharmaceutical industry. Heme can be used as the raw material of protoporphyrin drugs to produce protoporphyrin drugs for the treatment of various liver diseases. For example, disodium protoporphyrin has curative effect on various liver diseases. In addition, hematoporphyrin derivatives prepared with heme as a raw material are used for the treatment of human tumors as they can concentrate in tumors and have an enhanced response to ultraviolet lasers. So, when the patient is treated with red lasers, hematoporphyrins produce free radicals to kill tumor cells.

9.3 Carotenoids

Carotenoids, also known as polyene pigments, are widely distributed pigments in natural food materials. Red, yellow and orange fruits and vegetables are carotenoid-rich foods, and carotenoids are also found in animal materials such as egg yolk and shrimp shells. In general, chlorophyll-rich plant tissues are also rich in carotenoids because chloroplasts and chromoplasts are organelles with abundant carotenoid content.

The structure of carotenoids can be classified into two categories: ***carotenes*** contain only parent hydrocarbon chains; and ***xanthophylls*** contain oxygen as a functional group, such as hydroxyl, epoxy, aldehyde and ketone groups, on the parent hydrocarbon chains.

9.3.1 Carotene

9.3.1.1 Structure and Basic Properties

Carotenes include four compounds, namely ***α-carotene***, ***β-carotene***, ***γ-carotene*** and ***lycopene***, all of which are C_{40} tetraterpenoid pigments and biosynthesized by the linkage of two C_{20} geranylgeranyl diphosphate molecules.

It can be seen from Fig. 9.13 that they are closely related compounds with similar chemical properties, but their nutritional properties are different. For example, α-carotene, β-carotene and γ-carotene are provitamin A, which means they can be converted into vitamin A in the body. One molecule of β-carotene can be converted into two molecules of vitamin A, while each molecule of α-carotene or γ-carotene can only be converted into one molecule of vitamin A. Lycopene is not a provitamin
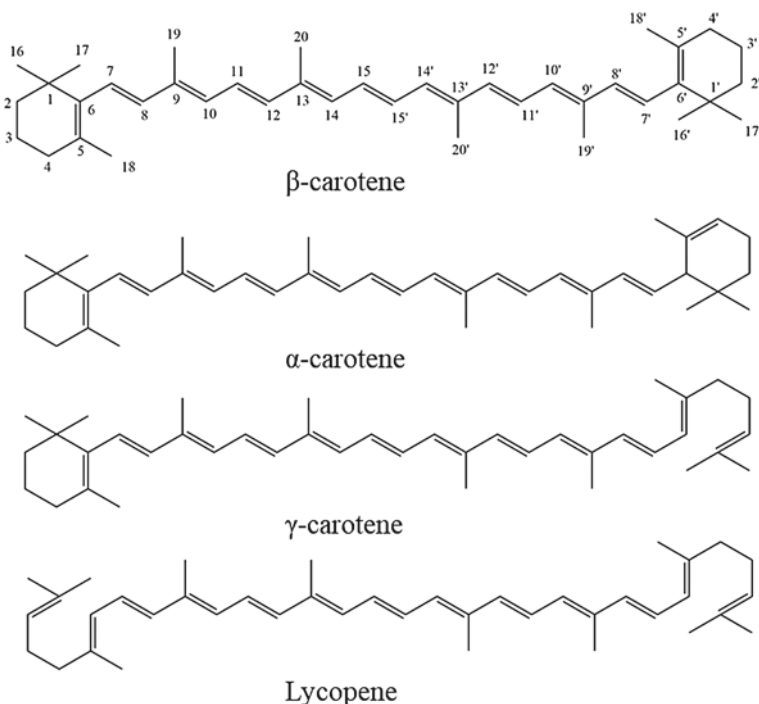
Fig. 9.13 Structures of carotene

A. Geometric isomers of carotene refer to geometric isomerization of one or more of their double bonds and the hydrides of carotenes refer to their hydrogenation products, such as phytoene.

α-Carotene and β-carotene are widely present in foods and biological materials, especially in carrots, sweet potatoes, egg yolks and milk. Lycopene is the main pigment of tomato and is also widely found in fruits such as watermelon, pumpkin, citrus, apricot and peach. In plant tissues, they are mainly found in chromoplasts; in animals, they are mainly distributed in specific tissues rich in lipids, such as egg yolk.

Carotene is a typical fat-soluble pigment, easily soluble in organic solvents such as petroleum ether and ether, and insoluble in ethanol and water. Since carotenes have many conjugated double bonds in their structures, they are easily oxidized and the resulting products are very complicated. Carotenes are susceptible to oxidation when plant tissues are damaged; lipoxygenase, polyphenol oxidase and peroxidase always accelerate the indirect oxidation of carotenes, as they first catalyze the oxidation of their respective substrate to form an intermediate which in turn oxidizes carotene. For example, lipoxygenase catalyzes the oxidation of unsaturated fatty acids to form hydroperoxides, which in turn react with carotenes. Therefore, in food processing,
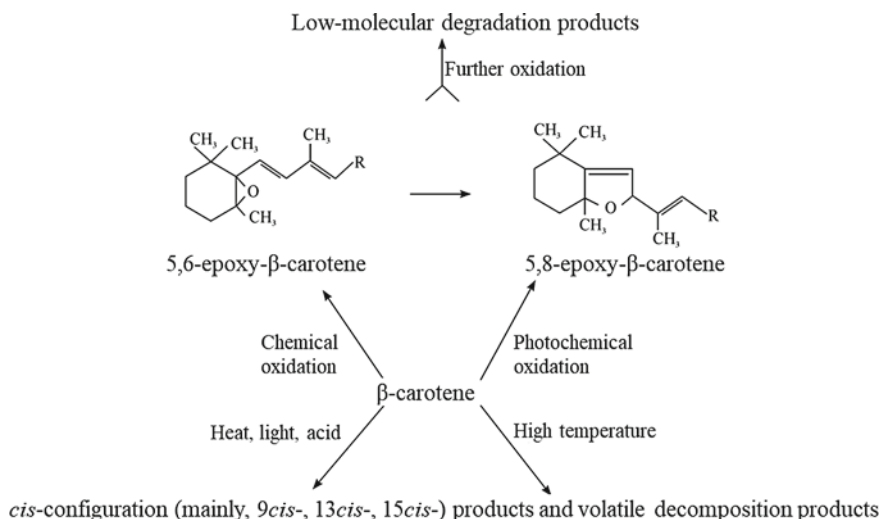
Fig. 9.14 β-carotene degradation reaction

appropriate treatment for inactivation enzyme such as blanching is helpful for the protection of carotene pigments.

In general, the conjugated double bonds of carotene are mostly in the all-trans configuration, and only a very small number of cis isomers are present. Carotenoids are highly susceptible to isomerization under heat treatment, organic solvents, acid and light conditions. Since carotene has many double bonds, there are many types of isomers, such as β-carotene, which have 272 possible isomers. Figures 9.14 summarizes the degradation reactions and possible isomerization reactions of β-carotene.

Since carotene is easily oxidized, it undoubtedly has a good antioxidant effect, which can scavenge singlet oxygen, hydroxyl free radicals, superoxide free radicals and peroxyl free radicals. When carotene exerts its antioxidant effect, it may be degraded or recovered after the antioxidant reaction.

9.3.1.2 Carotene Changes in Food Processing and Storage

In most fruit and vegetable processing, the properties of carotenoids are relatively stable. For example, freezing has very little effect on carotene pigments. However, under thermal processing conditions, when the plant tissue is heated, carotene is transferred from the colored tissue and dissolved in the lipid, so that its existing form and distribution in the plant tissue are changed, and it may be degraded under aerobic, acidic and heat conditions, as shown in Fig. 9.12. As provitamin A, the isomerization and degradation reactions of carotene in food during processing and storage sometimes are disastrous, which will definitely reduce the activity of provitamin A.

9.3.2 Xanthophyll

Xanthophyll pigments are widely found in biological materials, and they are more kinds of xanthophyll than carotene due to the addition of different oxygen groups. Some xanthophyll structures are shown in Fig. 9.15.

As the addition of oxygen group increases the hydrophilicity of xanthophyll pigments, they dissolve well in methanol or ethanol, but are less soluble in ether and petroleum ether. When extracting total carotenoids from plants, a compound solvent capable of extracting both carotene and xanthophyll should be taken into consideration, such as a proper ratio of hexane to acetone.

Xanthophyll is often yellow and orange, and a few are red, such as capsanthin. Like carotenes, xanthophylls are also prone to cis/trans isomerization under the action of heat, acid and light, and they are susceptible to degradation by oxidation and photooxidation, which sometimes change the color of the food and related flavor aspect. Some of the xanthophylls are also provitamin A, such as cryptoxanthin. Most of the xanthophyll are the same as carotene and have antioxidant effects.

9.3.3 Carotenoid Changes in Food Processing and Storage

Carotenoids in vegetables and fruits can be found with free form or in esterification with fatty acids, while esterification does not modify their chromophore properties of carotenoids but changes the chemical and biological properties. Some carotenoids are also combined with proteins. For example, scutellin (3,3′-dihydroxy-4,4′-diketone-β-carotene) combines with protein in a fresh lobster shell to form the typical blue color; when the lobster is cooked, the binding of the protein to scutellin is destroyed, and scutellin is oxidized to astaxanthin (3,3′,4,4′-tetraketone-β-carotene) with red color.

Carotenoid change in the procedure of fruit and vegetable processing depends on certain conditions. For example, freezing has little effect on carotenoid pigments. However, under heat, aerobic, light and acidic processing conditions, parts of carotenoids are transformed from trans to cis configuration, and further degraded due to oxidation and photooxidation reactions, etc. Some non-thermal processing techniques, such as high electric field pulse, high pressure and modified CO_2 package, may help to preserve carotenoids. Since carotenoids are easily oxidized, they have strong antioxidant properties to remove singlet oxygen, hydroxyl radical, superoxide radical and peroxyl radical. Thus, carotenoids are effective natural antioxidants in the food industry (Fig. 9.16).
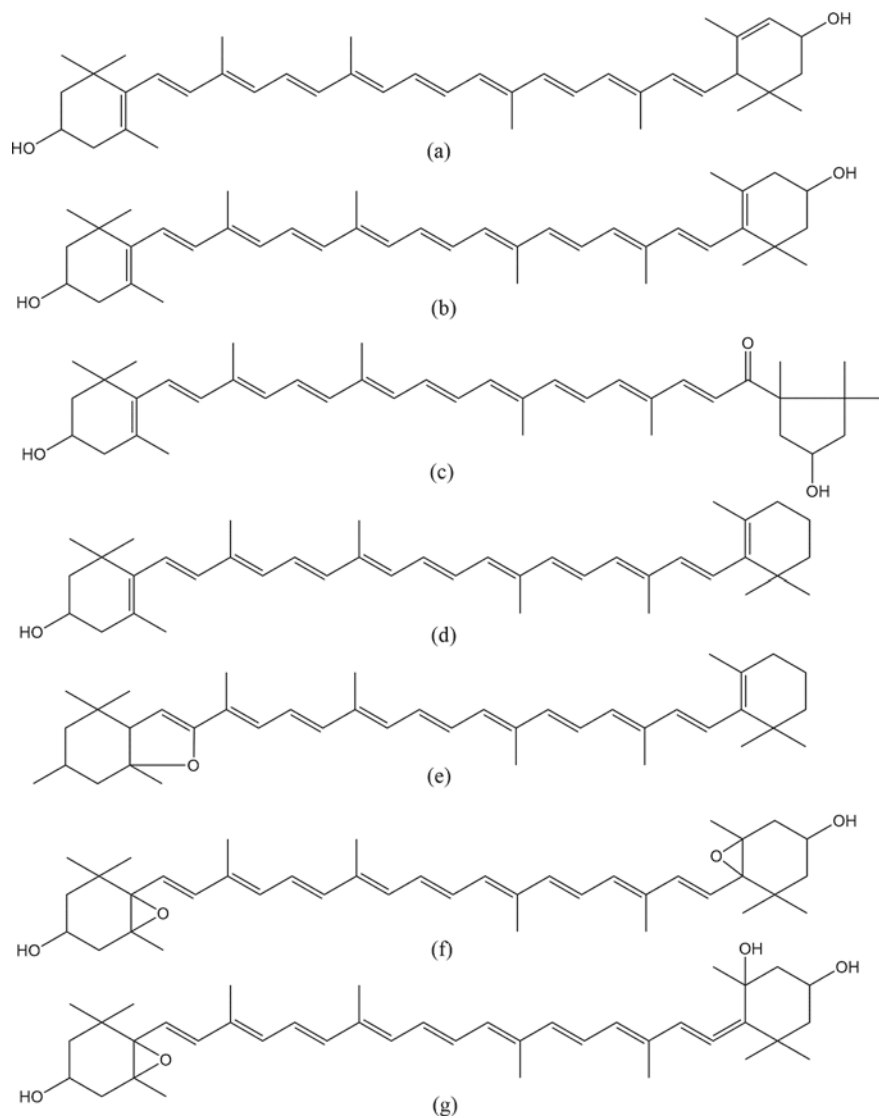
Fig. 9.15 Structures of some xanthophyll pigments that have been found in various plant materials as follows: **a** *lutein*, in marigold flower, orange, pumpkin and green leafy vegetable; **b** *zeaxanthin*, in wolfberry, maize, orange and red pepper; **c** *capsanthin*, in red pepper; **d** *cryptoxanthin*, in buah merah, papaya, mango, orange, maize, and persimmon; **e** *citroxanthin*, in orange; **f** *violaxanthin*, in mango, orange, jio and lamb's quarters; **g** *neoxanthin*, in spider wisp and chanca piedra

Fig. 9.16 The basic
structure (C6–C3–C6) of
flavonoid compounds
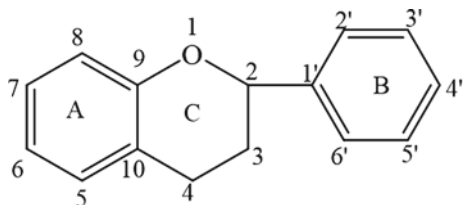
9.4 Polyphenolic Pigments

Polyphenols are a very broad class of compounds in nature with a basic core structure of **α-phenyl benzopyran**. Since two or more hydroxyl groups are attached to the benzene ring, they are collectively referred to as polyphenol pigments. Polyphenolic pigments are the main water-soluble pigments found in plants, and are mainly associated with flavonoid compounds.

9.4.1 Anthocyanins

In 1835, Marquart first extracted a blue pigment called anthocyan from the cornflower of chrysanthemum. The word cyan is taken from the Greeks Anthos (flowers) and Kyanos (blue). **Anthocyanins** are glycosides of **anthocyanidin**, a type of water-soluble pigment widely found in plants, so anthocyanidin is also called aglycone. Its colors include blue, purple, violet, magenta, red and orange, and is the substance that makes up the visual colors of many flowers and fruits.

9.4.1.1   Structure and Physical Properties

Anthocyanins have a carbon skeleton structure typical of flavonoids, and various anthocyanins and anthocyanidins are formed due to the difference in the number and type of substituents (Fig. 9.17). There are more than 20 anthocyanidins known, among which 6 are the most common anthocyanidins distributed in plant material, namely **cyanidin**, **pelargonidin**, **delphinidin**, **peonidin**, **petunidin** and **malvidin**. The sugars which are acylated with anthocyanidins are mainly glucose, galactose, xylose, arabinose and their disaccharides or trisaccharides. The aglycone sites of naturally occurring anthocyanins are mostly at the C_3 and C_5 positions of the 2-phenylbenzopyran cation, and a few at the C_7 position, or at the $C_{3'}$, $C_{4'}$ and $C_{5'}$ positions. These glycosyl groups are sometimes acylated by aliphatic or aromatic organic acids, and the main organic acids involved in the above reaction include caffeic acid, *p*-coumaric acid, sinapic acid, *p*-hydroxybenzoic acid, ferulic acid, malonic acid, malic acid, succinic acid or acetic acid.

Fig. 9.17 The structure of
anthocyanin
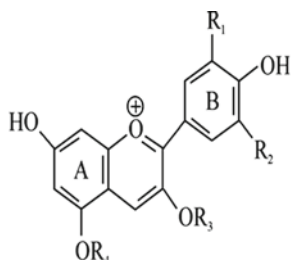
R_1, R_2= -H, -OH, or -OCH$_3$; R_3, R_4=Glycosyl, or –H

The difference in the color of various anthocyanins is mainly caused by the type and number of substituents. The substituents on the anthocyanin molecule are hydroxyl group, methoxy group or sugar group. As an auxochrome, the strength of the color-donating effect of the substituent depends on their electron-donating ability, and the stronger the electron-donating ability, the stronger the color-assisting effect. The electron-donating ability of the methoxy group, similar to the sugar group, is stronger than that of the hydroxyl group; the sugar group may exhibit a space hindrance effect due to the relatively large molecule. Figure 9.18 indicates that with
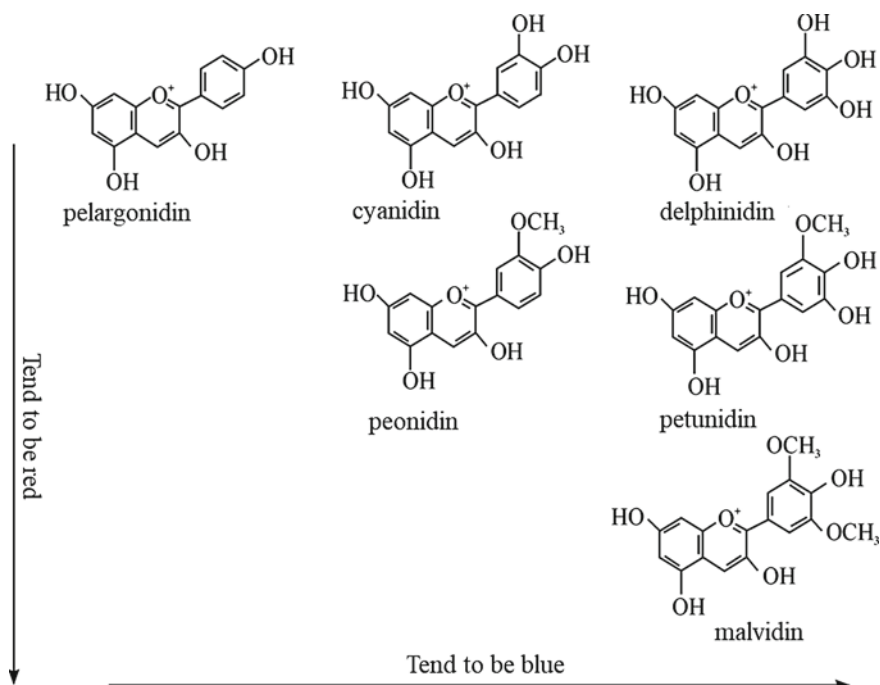
Fig. 9.18 The 6 types of common anthocyanidin in foods and their color changes

the increase of the –OH number, the light absorption wavelength shifts to the red light direction (red shift), and the blue color of anthocyanidin is strengthened; as the $-CH_3$ number increases, the light absorption wavelength shifts to the blue light direction (blue shift), and the red color is strengthened; due to red shift and blue shift, the color of anthocyanins deepens.

Both anthocyanins and anthocyanidins are water-soluble pigments, while anthocyanins have additional hydrophilic glycosyl groups, leading to their greater solubility in water.

More than 250 anthocyanins have been found in plants, and the variety of anthocyanins contained in plants varies with different growth stages and maturity periods, ranging from about 20 to 600 mg/100 g fresh weight.

9.4.1.2 Changes in Anthocyanins in the Food Industry

Anthocyanins and anthocyanidins are not stable, and they often change color due to chemical reactions during food processing and storage. Factors that affect their stability include pH, oxygen concentration, oxidants, nucleophiles, enzymes, metal ions and temperature.

The relationship between the structure of different anthocyanins and anthocyanidins and their stability have certain regularity. The stability of anthocyanins and anthocyanidins with more hydroxyl groups is not as high as those with more methoxy groups. Anthocyanins are not as stable as anthocyanidins, and the stability of different glycosyl groups is also different. For example, the color of plants with a high content of cyanidin, pelargonidin and delphinidin is not as stable as that of plants with a high content of petunidin and malvidin. Cranberry contains galactosyl anthocyanins, which are more stable during storage than arabinosyl anthocyanins.

1. Effects of pH

The solution pH significantly influences the color of anthocyanins due to the ionic nature of anthocyanin structures. In an acidic environment, some anthocyanins appear red, purple in natural pH while blue in alkaline conditions. When anthocyanins are native red, which are dominated by the structure of flavylium cations, they are quite stable in acidic solution, as the flavylium cation increases their solubility in water. When the solution pH increases, colorless structures such as alcohol pseudo base and chalcone are yielded, along with the formation of anionic quinonoidal structures which are blue due to hydration reactions of flavylium ion. Quinonoidal compounds are not stable at acidic conditions. If pH is between 4 and 5, anthocyanin solutions always show little color as a result of a few flavylium cations and quinonoidal anions (Figs. 9.19 and 9.20).

2. Temperatures

Temperature strongly affects the stability of anthocyanins and anthocyanins, and the extent of this effect is also affected by environmental oxygen content, anthocyanin species and pH conditions. In general, the thermostability of anthocyanins
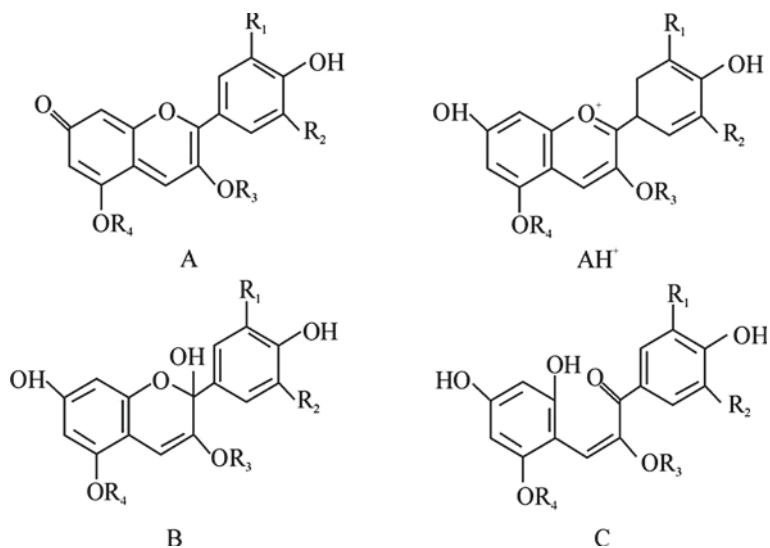
Fig. 9.19 The four forms of anthocyanins in aqueous solution and their colors. A is a quinone structure (blue), AH⁺ is a 2-phenylbenzopyran cation (red), B is an alcohol pseudo base structure (colorless) and C is a chalcone structure (colorless)
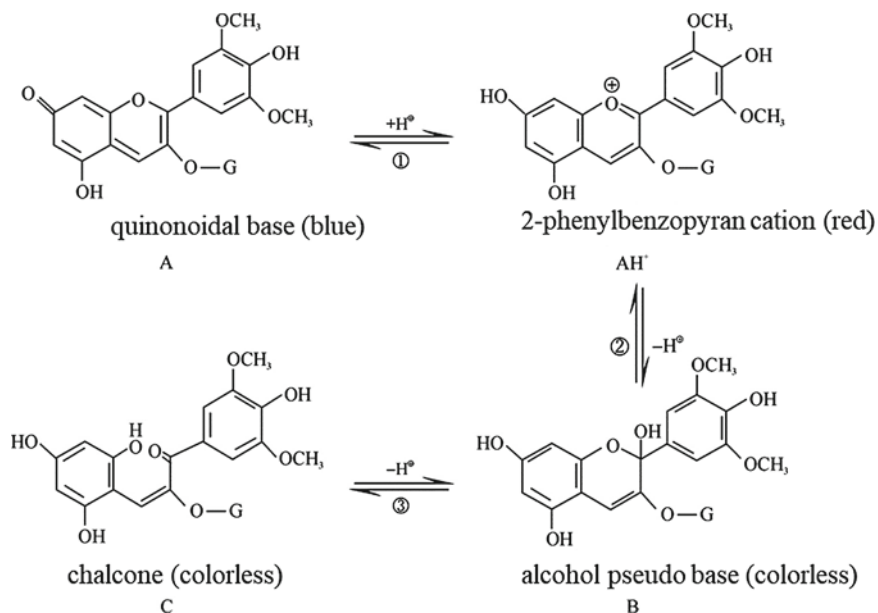

Fig. 9.20 Four kinds of structures with malvidin-3-glucoside appearing in the pH range from 0 to 6
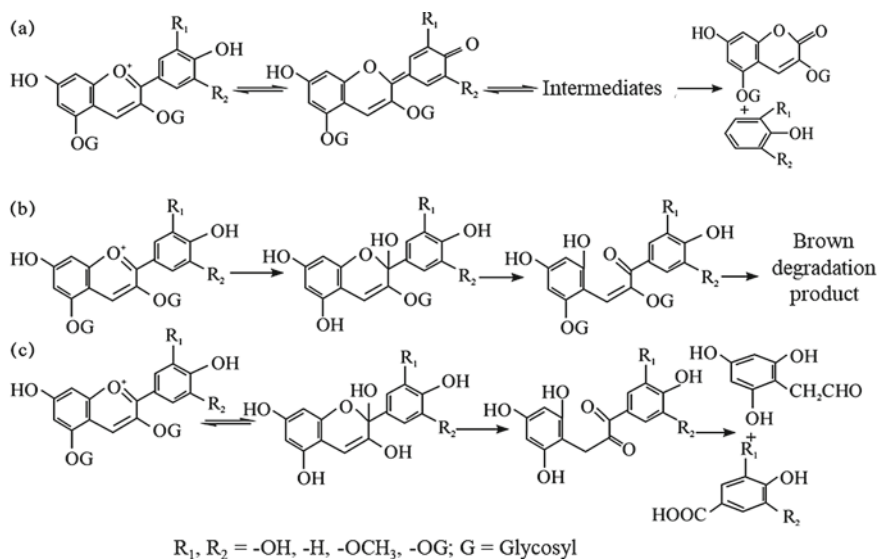
R₁, R₂ = -OH, -H, -OCH₃, -OG; G = Glycosyl

Fig. 9.21 Degradation mechanism of 3,5-diglucose-anthocyanins

and anthocyanidins containing more hydroxyl groups is not as stable as that with more methoxy or glycoside groups.

The conversion balance among the four structural forms of anthocyanins in an aqueous solution is also affected by temperature. When heating, the balance shifts toward the direction of the formation of chalcone structure, and the result is that the content of coloring substances (AH⁺ and A, Fig. 9.19) decreases. When cooled and acidified, the pseudo base anthocyanins are quickly converted to cation ones (AH⁺), while the chalcone type changes little.

The exact mechanism of thermal degradation of anthocyanins has not been fully elucidated, and three degradation pathways have been proposed (Fig. 9.21). 3,5-Diglucose-coumarin glycosides are common degradation products of 3,5-diglucose-anthocyanins. Route A (Fig. 9.21a) shows that this product is the first conversion of a 2-phenylbenzopyran cation to the quinone structure, and then the intermediate decomposes to produce coumarin derivatives and phenol compounds. In route B (Fig. 9.21b), the 2-phenylbenzopyran cation is first converted to a pseudo-basic structure, and then it decomposes into a brown degradation product through the chalcone structure. The first few steps of route C (Fig. 9.21c) are similar or identical to those of route B, but the degradation products of chalcone are formed by the insertion of water. These research results indicate that the thermal degradation of anthocyanins is affected by the types of anthocyanins and the degradation temperature. The higher the heating temperature, the faster the color change of anthocyanins, 110 °C is the most tolerable temperature for anthocyanins, and the decomposition rate of anthocyanins is low below 60 °C.

3. Oxygen, water activity and the effects of ascorbic acid

In the presence of oxygen, anthocyanins degrade to produce colorless or brownish substances due to their highly unsaturated structure which makes them sensitive to oxygen. If the grape juice is hot-filled and filled up totally, it takes relatively long time for the color of grape juice to change from purple to brown due to limited accesses of oxygen; if it is replaced by nitrogen filling or vacuum filling, the color change will be more slower. This indicates that oxygen has a destructive effect on anthocyanins or anthocyanidins.

There is not much research data on the influence mechanism of water activity on the stability of anthocyanins, but studies have confirmed that the stability of anthocyanins is relatively highest in the range of water activity from 0.63 to 0.79.

In juices containing ascorbic acid and anthocyanins, the levels of these two substances are reduced simultaneously. This is because ascorbic acid can produce H_2O_2 in oxidation, and H_2O_2 can do a nucleophilic attack on the C_2 of α-phenylbenzopyran cation, thereby splitting the pyran ring to produce a colorless ester and coumarin derivative. Further degradation or polymerization eventually produces a brown precipitate in the juice. Therefore, conditions for promoting or inhibiting oxidative degradation of ascorbic acid also work for anthocyanin degradation. For example, an increase of Cu^{2+} concentration will accelerate the degradation of ascorbic acid and anthocyanins, while ascorbic acid and anthocyanin are simultaneously protected in the presence of antioxidants such as quercetin in foods.

4. The influence of light

Light has a dual effect on anthocyanins. One is beneficial to the biosynthesis of anthocyanins, and the other is the degradation of anthocyanins. Under light conditions, acylated and methylated diglycosides are more stable than non-acylated diglycosides, and diglycosides are more stable than monoglycosides. After anthocyanins condense by themselves or with other organic substances, depending on the environmental conditions, the stability of anthocyanins may be increased or decreased. Polyhydroxyflavonoids, isoflavones, and auron sulfonates are resistant to the photodegradation of anthocyanins, as negatively charged sulfonic acid groups and positively charged 2-phenylbenzopyran cations attract each other, making these molecules form a complex with anthocyanins (Fig. 9.22).

Other radiation energy can also cause anthocyanin degradation. For example, when using ionizing radiation to preserve fruits and vegetables, there is photodegradation of anthocyanins.

5. Effect of sulfur dioxide

SO_2 is a preservative commonly used in the food industry. The decolorization of anthocyanin by sulfur dioxide is either reversible or irreversible. When the use of SO_2 is between 500 and 2000 µg/g, the color can be partially recovered by elution with a large amount of water in the subsequent processing. The irreversible bleaching effect suggests that the bleaching mechanism is that SO_2 forms bisulfite under the

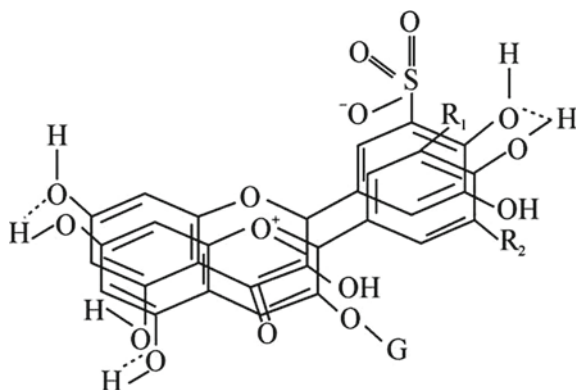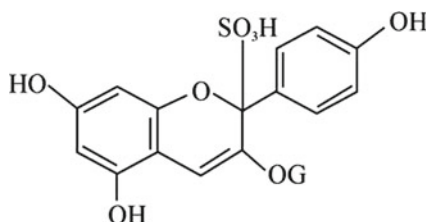**Fig. 9.22** Anthocyanin-polyhydroxyflavone sulfonyl complex

Fig. 9.23 Anthocyanin sulfite complex

acid condition in juice, and nucleophilic attacks occur on C2 or C4 of anthocyanin to produce colorless anthocyanin sulfite (Fig. 9.23).

6. Effects of sugar and sugar degradation products

When the sugar concentration is high, the color of the anthocyanin is well protected due to the low water activity. However, when the sugar concentration is low, i.e., in juice, the degradation or discoloration of anthocyanins is accelerated. These sugars first degrade (non-enzymatic browning) into furfural or hydroxymethyl furfural, and then react with anthocyanins to form a brown substance, which will be promoted by increasing temperature and oxygen concentration. This effect is more obvious when fructose, arabinose, lactose and sorbose are used than glucose, sucrose or maltose, which causes more sever problems in fruit juice production.

7. Metal ion

Anthocyanins can complex with metal ions such as Al^{3+}, Fe^{2+}, Fe^{3+}, Sn^{2+} and Ca^{2+}, thereby stabilizing the color of anthocyanins. However, this reaction can only occur when the anthocyanin contains vicinal hydroxyl groups on the B ring, and the product may be dark red, blue, green and brown (Fig. 9.24). Such metal ion complexes are common in plants. For example, the color of fresh flowers is brighter than anthocyanins as part of the anthocyanins in fresh flowers forms complexes with metal ions. The color of canned fruits and vegetables will be affected by the metal material. Once the paint on the inner wall of the tank does not qualify, the metal ions etched from
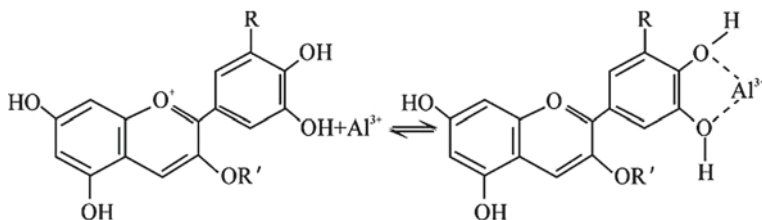
Fig. 9.24 Reactions between anthocyanins and metal ions

the inner wall of the tank often form complexes with anthocyanins, where in most cases this is undesirable, and in a few cases, the food color is beautified. It is advised to use stainless steel to process fruits and vegetables and apply citric acid to complex metal ions in food processing.

In the processing of peach, pear, lychee, cranberry and red cabbage, the problem of discoloration caused by the complexation of anthocyanin metal ions often occurs. The stability of this complex is higher than that of anthocyanin. Once it is formed, it is not easy to reverse, but citric acid can complex metal ions, which can reduce the formation of anthocyanin-metal complexes and can partially reverse them to anthocyanins.

8. Condensation of anthocyanins

Anthocyanins can undergo condensation reactions with themselves or other organic compounds to form weaker complexes such as proteins, tannins, other flavonoids and polysaccharides, by which the color of anthocyanins is modified by red shifting and the maximum absorption wavelength is increased. The formed pigments are also relatively stable during storage and processing. For example, the stable color in wine is partially due to the self-condensation of anthocyanins, and the polymer is insensitive to pH and somehow resistant to the bleaching of sulfur dioxide.

When 2-phenylbenzopyran cations and/or quinoid bases are adsorbed on a suitable substrate, such as glue or starch, the anthocyanins can be kept stable. When anthocyanins combine with certain nucleophilic compounds such as amino acids, phloroglucinol, catechol and ascorbic acid, they condense to produce a colorless substance, as shown in Fig. 9.25.

9. Hydrolysis of anthocyanins

The hydrolysis of anthocyanins includes acid hydrolysis and enzymatic hydrolysis. Generally, in a 1 mol/L HCl solution at 100 °C, anthocyanins are completely hydrolyzed to form corresponding anthocyanidins and sugars within 1 h. The higher the acidity, the faster the hydrolysis. Glucosidase and polyphenol oxidase can cause anthocyanin degradation. The former hydrolyzes the glyosidic bonds on the anthocyanin molecule to produce anthocyanidins and sugars; the latter catalyzes the oxidation of small molecular phenols to form orthoquinones, which can convert anthocyanins into oxidized anthocyanins and their degradation products by chemical oxidation.
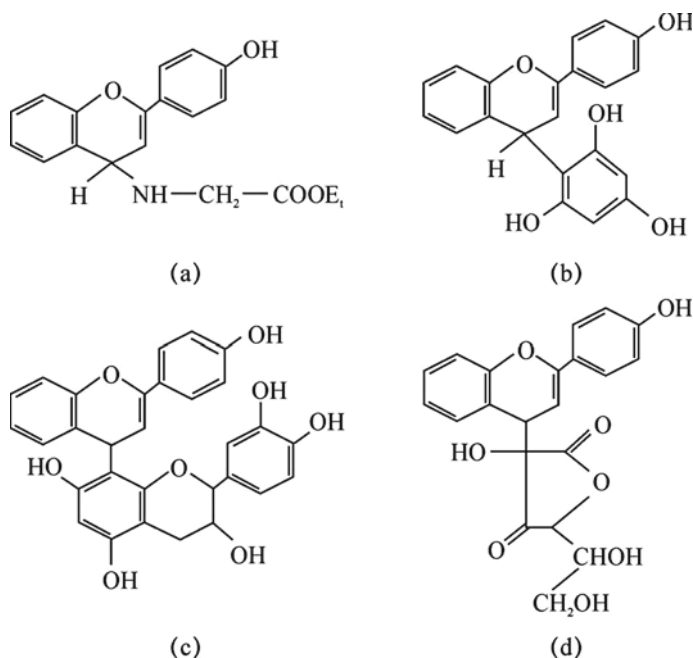
Fig. 9.25 The colorless condensate formed by 2-phenylbenzopyran cation with ethyl glycinate (**a**), phloroglucinol (**b**), catechin (**c**) and ascorbic acid (**d**)

Before processing, storage and packaging, preliminary steam bleaching can destroy and inhibit the anthocyanidase in fruits and vegetables. Glucose, gluconate and glucose delta-lactone are competitive inhibitors of glycosidase, and the activity of polyphenol oxidase can also be effectively inhibited by sulfur dioxide and sulfite. Therefore, in the process of fruit and vegetable processing, proper steam heating and the addition of enzyme inhibitors can effectively inhibit enzyme activity and protect the color of fruits and vegetables.

9.4.2 Flavonoid Pigments

9.4.2.1 Structure and Physical Properties

As shown in Fig. 9.8, flavonoids have the basic structure of C_6–C_3–C_6 framework, including flavonoid glycosides and free flavonoid aglycones in plant tissues. In flowers, leaves and fruits, most of them exist in the form of glycosides, while in xylem tissues, they are mostly in the form of free aglycones. The flavonoid compounds are further divided into the following subclasses as shown in Fig. 9.26 with their respective structures.
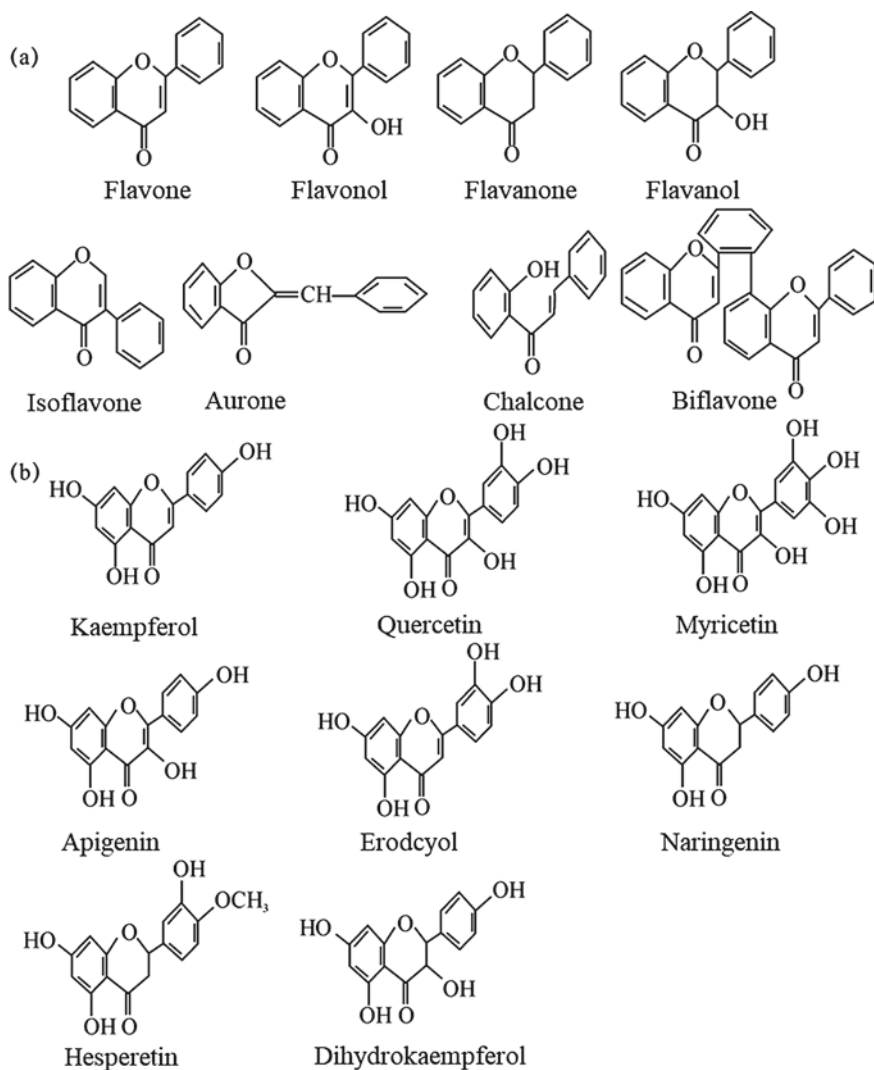
Fig. 9.26 Flavonoid compounds and their related structures. **a** The names and structures of some subtypes of flavonoids; **b** The names and structures of some common flavonoid compounds

It can be seen from Fig. 9.26 that flavonoids contain a wide range of natural pigments. Among these subclasses, anthocyanin has formed an independent category and has been introduced earlier in this chapter, and also, catechin belonging to flavanol will be discussed later. Flavonoids are also involved in the formation of another important phytochemical in vascular and non-vascular tissues, tannin. All these typical and important flavonoid-related pigments in the food industry will be discussed in detail in the following part.

Table 9.4 The effect of groups at different carbon positions on the color of flavonoids

C position	Group	Color
3	– OH	Only gray-yellow
3′ or 4′	– OH or –OCH$_3$	Mostly dark yellow

Flavonoids have many special biological functions such as anti-oxidation, anti-tumor, anti-mutation and cardiovascular protection. Besides anthocyanin with blue, red or purple color, other colored flavonoids are generally found in flavonoids. Flavonols, isoflavones, chalcone and flavonoid glycosides are mostly yellow. And in these subclasses, the number and binding position of phenolic hydroxyl groups in the structure of flavonoids have a great influence on their coloration. If only hydroxyl groups are present at the C_3 positions, the flavonoids are only gray-yellow; if there are hydroxyl groups at the $C_{3'}$ or $C_{4'}$ position, the flavonoids of the methoxy group are mostly dark yellow, and the hydroxyl group at the C_3 carbon position can strengthen the color of the compound having a hydroxyl group at the $C_{3'}$ or $C_{4'}$ carbon position (Table 9.4).

Natural flavonoids are mostly in the form of glycosides; flavonoid glycosides are easily soluble in water, methanol and ethanol solutions, and are insoluble in organic solvents. The glycosyl group of flavonoid glycosides is often glucose, galactose, xylose, rutinose, neohesperidose and gluconic acid. There are changes in the position of glyosidic bonds, but the most common glycosides occur at the C_7, C_5 and C_3 positions of the parent nucleus structure. There are also acyl substitutions in flavonoid compounds. There are more than 1,670 known flavonoids including their glycosides, and more than 400 are colored substances, most of which are pale yellow, and a few are orange-yellow.

Some flavonoids have a certain contribution to the color of food, but due to their light color, their contribution is small when the concentration is low. The light-yellow color of cauliflower, onion and potato is mainly produced by flavonoids. Like anthocyanins, flavonoids can also form condensates. After condensation, the color changes. The condensed flavonoids in cauliflower, onion and potato are considered as an important substance for the visible color of these vegetables.

9.4.2.2 Flavonoid Changes in Food Processing and Storage

Flavonoids form complexes with a variety of metal ions, and these complexes have a stronger coloration effect. For example, the complexation of flavonoids with Al^{3+} enhances yellow color. The maximum absorption wavelength of erodcyol and Al^{3+} is 390 nm, which is very attractive. The flavonoids can be blue, purple, brown and black after complexation with iron ions; and the rutin (3-rutinosyl-quercetin) in asparagus produces an unsightly dark color when it encounters iron ions, causing dark spots in asparagus. On the contrary, when rutin is complexed with tin ions, it produces an ideal yellow color.
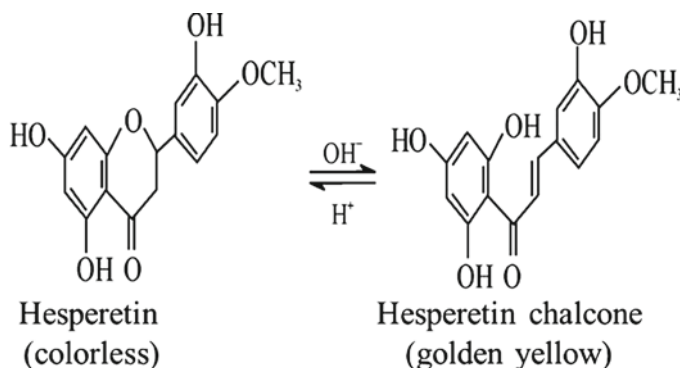
Fig. 9.27 Colorless flavanones are heated with alkali to transform into colored chalcone

In food processing, sometimes the pH is increased due to the use of sodium carbonate and sodium bicarbonate, or the high hardness of the water. Under such conditions, the originally colorless flavanone or flavanol can be converted into colored chalcone (Fig. 9.27). For example, potatoes, wheat flour, alfalfa, onion, cauliflower and kale will turn from white to yellow when processed (cooked) in alkaline water. This change is a reversible change that can be controlled and reversed with an organic acid.

The ethanol solution of flavonoids rapidly appeared red or purple under the reducing action of magnesium powder and concentrated hydrochloric acid. For example, flavonoids turn orange-red, flavonols turn red, and flavanones and flavanols turn purple-red. This is due to various anthocyanins that are formed after the reduction of flavonoids.

Flavonoids are also polyphenols. The intermediate products of enzymatic browning such as o-quinone or other oxidants can oxidize flavonoids to produce brown precipitates. The black color of mature olives is formed by the oxidation of erodcyol-7-glucoside during product fermentation and later storage; it is also one of the reasons for the long-term browning of the juice and the precipitation.

9.4.3 Catechins

Catechin, also called tea polyphenol, with the name derived from catechu of *Acacia catechu* L. extract, is 3,3′,4′,5,7-pentahydroxyflavan with two steric forms of (+)-catechin. In addition, catechin also contains its derivatives showing a similar molecular arrangement. There are six common catechins in tea, namely L-epigallocatechin, L-gallocatechin, L-epicatechin, L-Catechin, L-Epicatechin gallate and L-Epigallocatechin gallate (Fig. 9.28).

Catechins are widely distributed in food materials and herbs including tea, persimmons, apples, cacaos, berries and grapes. Catechins are high in tea. Catechin itself is
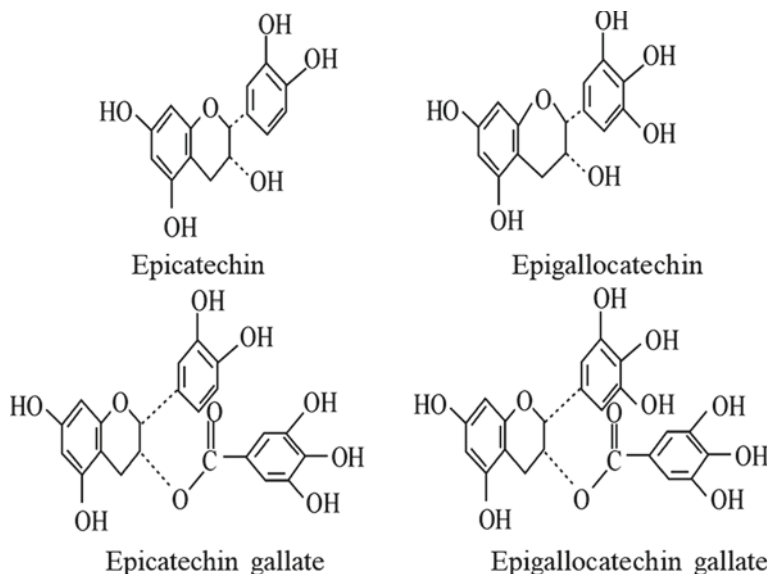
Fig. 9.28 The structure of several common catechins

colorless and has a slight astringency, while the combination of catechin and metal ions produces a white or colored precipitate. For example, a catechin solution reacts with ferric chloride to form a black-green precipitate, and a yellow precipitate is formed in the presence of lead acetate.

As a polyphenol, catechins are very easily oxidized to form a brown substance. Many catechin-containing plant tissues also contain polyphenol oxidase and/or peroxidase. When the tissue is damaged, catechins are oxidized by these enzymes to form a brown substance. The intermediate product of enzymatic browning, ortho-quinone, is an important substance that causes further oxidation of catechins or oxidative polymerization (Fig. 9.29). In the processing of black tea, the oxidation products of catechins are oxidized to form theaflavins and thearubigins, which help to form the color of black tea. Catechins can also be automatically oxidized when exposed to oxygen under high temperature and humid conditions.

9.4.4 Tannin

Tannin is an important kind of polyphenolic component with over 160 000 tons potentially biosynthesized each year in the world. It can be found not only in all vascular plant tissues but also in some non-vascular plants, i.e., marine algae. In food materials, tannins are extremely high in gallnuts and persimmons. Traditionally, tannins are divided into two types: **hydrolysable** and **condensed tannins** (or
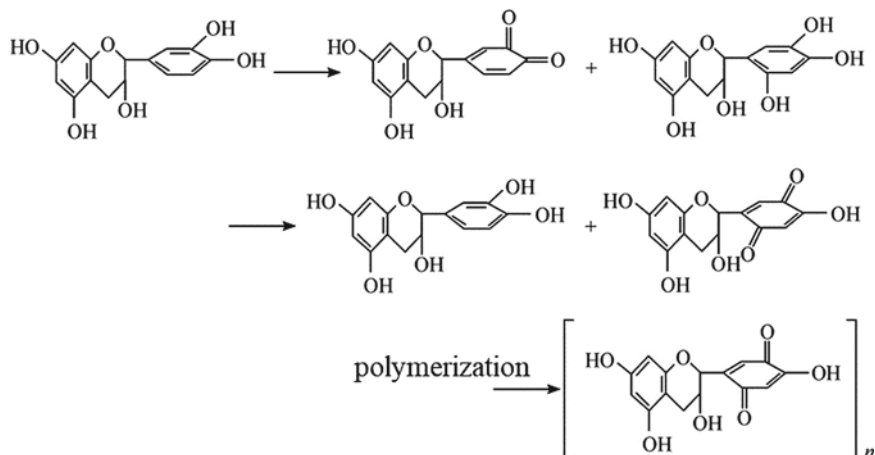
Fig. 9.29 The color change of catechin

anthocyanogen). The hydrolysable tannin molecule is composed of phenols and sugar (i.e., glucose) esters. According to the phenol type, hydrolysable tannins are further classified into two families, **gallotannins** that yield gallic acids and related derivatives after hydrolysis, and **ellagitannins** that produce ellagic acids and their derivatives.

The basic structural unit of condensed tannins is a flavan-3-ol or flavan-3,4-diol, and 2–8 of flavonoid repetitions are needed to form condensed tannins, which are generally in a complex with proteins and were first found in cocoa beans and later found to be ubiquitous in fruit juices, and now condensed tannins occupy more than 90% of worldwide tannin production. Besides hydrolysable and condensed tannins, another complex tannin should be also considered. **Complex tannins** are always formed by ellagitannin unit and flavan-3-ol unit with a typical representative as acutissimin A shown in Fig. 9.30, where the flavagallonyl group connects to a polyol derived from D-glucose by a glucosidic connection in C_1 and three other ester bonds.

Condensed tannins are converted to anthocyanins and catechins under acidic heating conditions, such as pelargonidin, petunidin and delphinidin. For example, the dimeric anthocyanogen in apples, pears and other juices can be converted into anthocyanidins and other polyphenols when heated under acidic conditions. The mechanism of this reaction is shown in Fig. 9.31.

Condensed tannins also produce oxidation products during processing and storage. For example, when the juice is exposed to the air or under light, they turn into a stable reddish-brown substance, which is responsible for the discoloration of apple juice. It is generally believed that the intermediates of enzymatic browning can also oxidize condensed tannins.

The color of tannin is yellow or slightly brown, which has a strong astringent taste, and they can precipitate with proteins and combine with various alkaloids or polyvalent metal ions to form colored insoluble precipitates. In food storage,
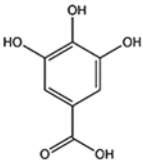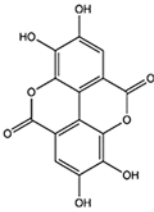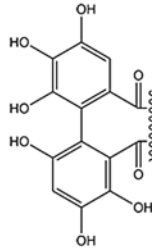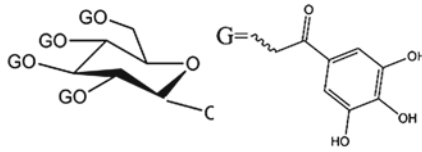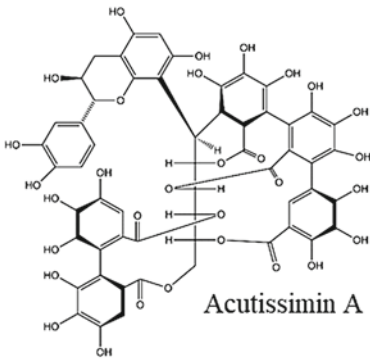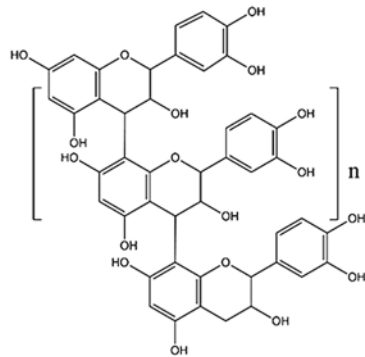
Fig. 9.30 Tannin classifications and their respective examples

tannins condense under certain conditions (such as heating and oxidation), thereby eliminating astringency. As polyphenols, tannins are also susceptible to oxidation, both enzymatic browning and non-enzymatic browning, and the former is dominant.
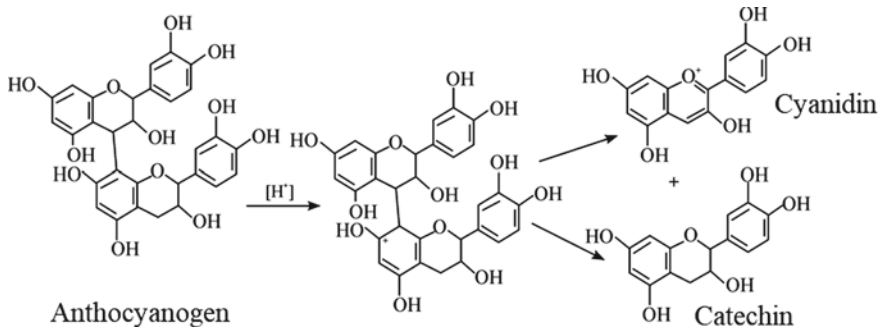
Fig. 9.31 The mechanism of acid hydrolysis of anthocyanogen

9.5 Food Colorant

9.5.1 Caramel Color

Caramel color (caramel) is a complex red-brown or dark-brown mixture formed by dehydration and condensation of carbohydrate raw materials, such as cerealose, sucrose, molasses, invert sugar, lactose, maltose syrup and starch hydrolysate, during the heating process. And it is a semi-natural food coloring agent that is widely used. According to the different catalysts used in the production process of caramel coloring, the Codex Alimentarius Commission (CAC) divides it into four categories (Table 9.5).

Caramel pigment is a dark brown gel or lump, with a special sweet aroma and pleasant bitterness, but it is rarely displayed under normal usage. It is easily soluble in water and has good stability to light and heat.

Caramel pigments have colloidal properties and are charged. The type of charge is related to the production process of caramel and the pH environment of the food. Therefore, when choosing a caramel colorant, it is necessary to consider that the charge of the caramel should be the same as that of the food, otherwise flocculation or precipitation will occur. For example, the caramel pigment added to beverages should have a strong negative charge, and the isoelectric point should be less than 1.5, and its pH range is mostly between 2.5 and 3.5, while the caramel pigment added to soy sauce and beer should usually be positively charged, and the pH range should be from 3.8 to 5.

The caramel pigment produced by the ammonia method currently occupies the largest market for caramel pigment. This type of caramel pigment may contain 4-methylimidazole, which is a convulsant. The results of chronic toxicity tests have confirmed that it will decrease the number of leukocytes, with slow growth. Therefore, in the caramel pigment produced by the ammonium salt method, the content of 4-methylimidazole must be strictly controlled.

Table 9.5 Types and characteristics of caramel colorant

Characteristics	Caramel type			
	Ordinary caramel (I)	Sulfite caramel (II)	Ammonia caramel (III)	Ammonium sulfite caramel (IV)
International code	ISN 150a	ISN 150b	ISN 150c	ISN 150d
	EEC No. E150a	EEC No. E150b	EEC No. E150c	EEC No. E150d
Typical use	Distilled alcohol, sweets, etc.	Alcohol	Baked goods, beer and soy sauce	Soft drinks, soup, etc.
Electric charge	Negative	Negative	Positive	Negative
Whether it contains ammonia compounds	No	No	Yes	Yes
Whether it contains sulfur compounds	No	Yes	No	Yes

Note ISN is the international numbering system for food additives adopted by the International Codex Alimentarius Commission (CAC) in 1989 (revised in 2001) and EEC is the European Community

According to the provisions of FAO/WHO, caramel pigment can be used in orange peel jelly, broth, cold drinks and other foods, and the dosage can be determined according to the normal production requirements.

9.5.2 Monascin Pigment

Monascin is derived from microorganisms. It is produced from a group of *Monascus* sp., *Monascus purpureus*, *Monascus anka* and *Monascus barkeri*, and it belongs to ketone pigments. There are six kinds of pigments, namely rubropunctamine, monascorubramine, rubropunctatin, monascorubrin, monascine and ankaflavine. There are red, yellow or purple pigments. Some of their structures are shown in Fig. 9.32.

The composition of monascin pigment obtained from different strains is different. For example, what is obtained from *Monascus purpureus* is ankaflavine, and what is obtained from *Monascus purpureus* is monascine. The physical and chemical properties of the above six monascin pigments are different from each other. The main ones with practical application value are rubropunctamine and monascorubramine.

Monascin pigment is a red or dark red powder, or liquid paste. The melting point is about 60 °C, and they are soluble in ethanol aqueous solution, ethanol, ether and glacial acetic acid. The color does not change with the pH value, and the
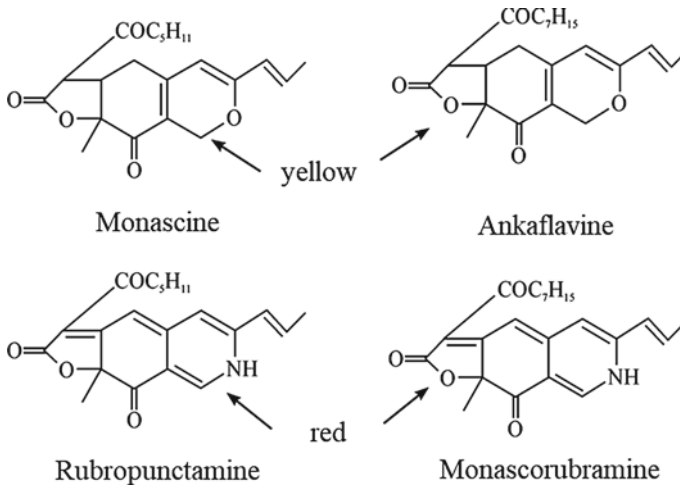
Fig. 9.32 The structure of monascin pigment

thermal stability is high. It is hardly affected by metal ions (such as Ca^{2+}, Mg^{2+}, Fe^{2+} and Cu^{2+}), and is also hardly affected by oxidants and reducing agents (except hypochlorous acid). However, under direct sunlight, the chromaticity is reduced. It has a good dye effect on protein. Once the protein is dyed, it will not fade after washing.

Monascin pigment also has an antiseptic effect, and has a strong inhibitory effect on *Bacillus cereus*, *Bacillus subtilis* and *Staphylococcus aureus*; secondly, it also has a certain inhibitory effect on *Pseudomonas aeruginosa*, *Pullorum* and *Escherichia coli proteus*; it has no inhibitory effect on *Sarcina*, *Saccharomyces cerevisiae* and *Penicillium chrysogenum*. The combination of monascin pigment, nisin and potassium sorbate can inhibit the growth of *Clostridium botulinum*. In addition, monascin pigment also has health effects such as reducing triglycerides, cholesterol and preventing arteriosclerosis. Monascin pigment can be used for the coloring of meat products. For example, the use of a coloring agent for fermented sausages can partially replace the amount of sodium nitrite. The color of fermented sausages made with 1 600 mg/kg monascin pigment as the coloring agent is close to that with 150 mg/kg sodium nitrite, and it has a certain inhibitory effect on *Clostridium botulinum*. Monascin pigment is used for the coloring of red fermented bean curd food, which not only provides the desired color of red preserved bean curd, a variety of aromas and fragrance components, but also has the functions of lowering blood pressure and cholesterol. Monascin pigment can also be used in the preparation of wine, fruit vinegar, beverages, various condiments, brewed foods, vegetable protein foods, etc.

Fig. 9.33 The structure of curcumin
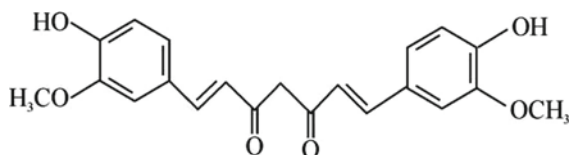
9.5.3 Curcumin

Curcumin, or turmeric yellow, is a yellow pigment extracted from the underground rhizomes of *Curcuma longa* in the ginger family. It is a group of ketone pigments. The main components are curcumin, demethoxycurcumin and bisdemethoxycurcumin. Its core structure is shown in Fig. 9.33.

Curcumin is an orange-yellow crystalline powder, almost insoluble in water, but soluble in ethanol, propylene glycol, glacial acetic acid and alkali solutions or ethers. It has a special fragrance, is slightly bitter, and yellow in neutral and acidic solutions, and brownish red in alkaline solutions. Curcumin is unstable to light, heat, oxidation and iron ions, but has good resistance to reduction. It has good coloring power to protein and is often used for coloring curry powder. Curcumin is also used in various oils to restore the color lost during processing.

9.5.4 Betalain

Betalain is a group of water-soluble pigments extracted from the tubers of the red beet from *Liliaceae*, and it is also widely present in flowers and fruits. It is present in these plants in the form of betacyanin and betaxanthin and their glycosides in the vacuole and the structures are shown in Fig. 9.34.

Betacyanin solution is purple-red in the pH range of 4–7. When the pH value is lower than 4 or higher than 7, the color changes to purple. When the pH value is above 10.0, betacyanin is hydrolyzed to betaxanthin. The solution immediately turned yellow. The heat resistance of betalain is not high, and it is relatively stable at pH 4.0–5.0. It will be converted into betalamic acid (BA) and cyclodopa-5-O-glucoside (CDG), and the reaction is reversible when the pH drops between 4 and 5.

Betanin can cause isomerization under the action of heating and acid, and two epimers can be formed in the chiral center of C_{15}. As the temperature rises, the proportion of isobetanin increases (Fig. 9.35), causing serious fading.

Betalain is also not resistant to oxidation, and the mechanism of oxidation is not clear, but it is not a free radical mechanism. For example, bleaching powder or sodium hypochlorite can make it fade, and the oxygen in the headspace of canned beets will speed up the fading of betalain. Light will accelerate oxidation, and ascorbic acid can slow down oxidation. The purple-red color fades after betalain is oxidized, and
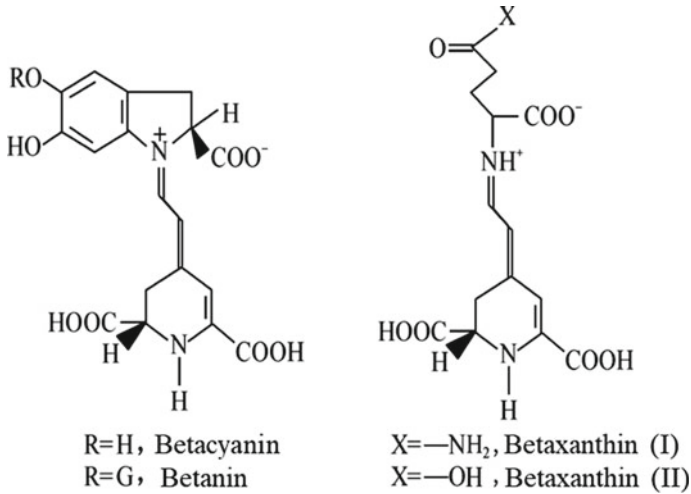
Fig. 9.34 The structure of betacyanin and betaxanthin
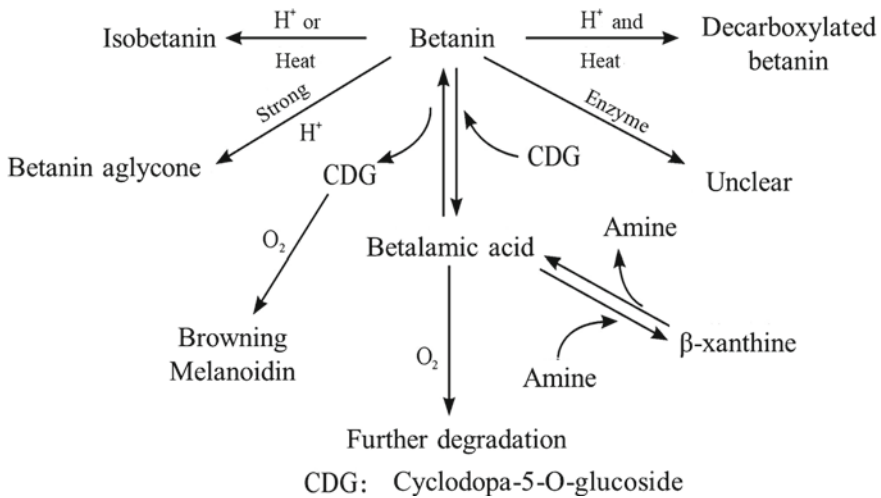
Fig. 9.35 Acid and/or thermal degradation of betanin

browness is often produced at the same time. If there is no oxidation condition, the stability of betalain to light is still good.

Certain metal ions also have a certain effect on the stability of betalain, such as Fe^{2+}, Cu^{2+} and Mn^{2+}. The mechanism is that these metal ions can catalyze the oxidation of ascorbic acid, thereby reducing the protective effect of ascorbic acid on betalain. The presence of metal chelating agents can greatly improve the effect of ascorbic acid as a betalain protector.

Betalain has good food coloring properties. The color is stable when used in foods with a pH from 3.0 to 7.0. In foods with low water activity, the color can be maintained for a long time.

9.5.5 Other Natural Colorants

Countries around the world also allow the use of a variety of other natural colorants, such as safflower yellow, shellac red, bilberry red, chili red, red-rice red, black-currant red, mulberry red, natural amaranth red, vine spinach red, black bean red, sorghum red, radish red, gardenia yellow, chrysanthemum yellow, corn yellow, *Hippophae rhamnoides* yellow, cocoa-shell pigment, tanoak brown, *Rosa laevigata michx* brown and *Quercus* brown.

9.6 The Principle and Practical Application of Food Toning

9.6.1 Preparation of Colorant Solution

The colorant powder is inconvenient to use directly, it is unevenly distributed in the food and may form pigment spots, and often needs to be formulated into a solution for use. The synthetic colorant solution generally uses a concentration of 1–10%, and it is difficult to adjust the hue if the concentration is too large.

In preparation, the weighing of the colorant must be accurate. In addition, it should be prepared according to the amount of each time, as the prepared solution is easy to precipitate after a long time. Due to the influence of temperature on the solubility of the colorant, the concentrated solution of the colorant that is prepared in summer will precipitate when stored in the refrigerator or during the winter. The aqueous solution of carmine turns black after long-term storage.

The water used in the preparation of the colorant aqueous solution should usually be boiled and cooled before use, or distilled water or the water treated with ion exchange resin should be used.

When preparing the solution, it should be avoided using metal utensils as the colorant may interact with metals and show different visual color; and in the storage, it should be avoided direct sunlight, preferably in a cool and dark place.
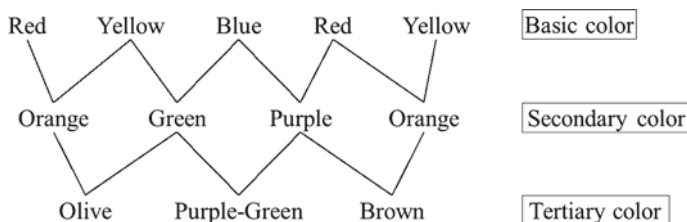
9.6.2 Principles of Hue Selection for Food Coloring

Hue is an attribute of visual perception that a surface presents one or two colors like red, yellow, green and blue. Most foods have rich colors, and their hue is closely

related to the inner quality and outer aesthetic characteristics of the food. Therefore, in the production of food, what hue is used in the food is very important. The choice of food color is based on the psychological or customary requirements for food color, and the relationship between color, flavor and nutrition. The hue selection should be like the original color of the food or consistent with the name of the food, and the color matching principle should be used to formulate the corresponding characteristic color of the specific food. For example, canned cherries and bayberry jam should choose corresponding cherry red and bayberry red. Red wine should choose purple, and brandy, yellow–brown. Another example is that the color of candy can be selected according to its flavor characteristics. For instance, mint candies tend to be green; orange candies, red or orange; chocolate candies, brown, etc.

9.6.3 Hue Tuning

With red, yellow and blue as the basic colors, two or three of them can be selected to form a variety of different color spectrums according to different needs. The basic method is to combine the basic colors into secondary colors, or combine them into tertiary colors. The simple tuning principle is as follows.

Red	Yellow	Blue	Red	Yellow	Basic color
Orange	Green	Purple	Orange		Secondary color
Olive	Purple-Green	Brown			Tertiary color

A variety of food synthetic colorants dissolved in different solvents can produce different hues and color intensities, especially when two or more food synthetic colorants are used for color matching, the situation effect is more obvious. For example, a certain proportion of a mixture of red, yellow and blue is yellower in aqueous solution, but redder in 50% ethanol. Due to the different alcohol content in food and aqueous solution, the hue after the colorant is dissolved is also different, so it is necessary to match colors according to the alcohol content and the intensity of the colorant. In addition, the food is moist when it is colored. When the moisture evaporates and gradually dries, the colorant will also concentrate on the surface layer, causing the so-called "concentration effect", especially when the affinity between the food and the coloring agent is low. When matching colors, pay attention to the different stability of various pigments, which will lead to changes in the hue of the synthetic color. For example, indigo fades faster, while lemon yellow is not easy to fade, so the initial matched green will gradually change to yellow-green. Synthetic pigments use the above principles to match the color. Natural pigments are not easy

to match colors due to their low adherence, easy discoloration and strong sensitivity to the environment.

9.7   Summary

Food pigments refer to substances in food that absorb and reflect visible light waves and then present foods in various colors. There are various methods for classifying food pigments. According to their sources, food colorants are classified into two categories: natural food colorants and synthetic food colorants; natural food pigments are further classified into plant pigments, animal pigments and microbial pigments according to their origins. According to their solubility, food pigments are classified into water-soluble and fat-soluble ones. Based on the chemical nature of food natural pigments, they are mainly divided into tetrapyrrole, isoprene and polyphenol categories, and their representatives were discussed in this chapter. Changes in natural pigments during food processing such as chlorophyll, heme, carotenoids and flavonoid pigments are the basis for food color changes. Understanding their reaction mechanisms is important to control their changes and ensure food color.

Questions

1. What are food pigments? What is their role in the food industry?
2. Under what conditions are the main chlorophyll derivatives produced? How to control the conditions in food storage and processing to keep food green?
3. What chemical changes can occur when meat is cured? Does excessive colorant cause any hazardous results?
4. Carotenoid pigments are multi-functional natural pigments, what about their nutritional effects?
5. What substances are included in polyphenol pigments? What are the factors that affect the color change of polyphenol pigments?
6. What are the main similarities and differences between natural colorants and synthetic colorants? How to choose food colorants for color matching?
7. What kind of color for food will be liked by people? Will the color of these foods change during processing and storage?
8. Why do shrimp turn from cyan to red after high-temperature processing?
9. Why does the surface color of meat change from purple-red to bright red, and again to brown, if it is placed at room temperature for a long time?
10. Why does the color of purely fermented wine change from purple-red to blue after adding baking soda?
11. What is the difference between synthetic pigments and natural pigments? Why are synthetic pigments replaced by natural pigments widely used in foods?
12. Explain the following terms: chlorophyll, heme, carotenoids, anthocyanins, metmyoglobin, oxymyoglobin and flavonoids.

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