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# CAROTENOIDS AS NATURAL COLORANT : A REVIEW

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## ABSTRACT

Color is quality attribute that is usually used by consumer as first assessment to choose the food product. However, food processing is one of process which can degrade the food's color, so the colorant is usually added. On this era, consumer tend to choose food products that have functional benefit. One of natural colorant which has it is carotenoids. Carotenoids gives red, orange, and yellowish. Carotenoids are divided into two groups, carotene and xanthophyll. Carotene consists of α-carotene, β-carotene, γ-carotene, and lycopene. Meanwhile, xanthophyll consists of β-cryptoxanthin, lutein, zeaxanthin, astaxanthin, fucoxanthin, and peridinin. This pigment is lipophilic so it can dissolve in oils and organics solvents and is quite resistant to heating, however it can be very easily degraded in acidic, light, and oxygen condition. Beside act as colorant, this pigment can act as antioxidant and provitamin A. The source of carotenoids is widely spread in flowers, fruits, tubers, leaves, and fruit peels. Extraction of this pigment can be done in three ways, there are maceration extraction, supercritical fluid extraction, and enzymatic extraction.

**Keywords**: Antioxidant, extraction, carotenoids, food colorant

## INTRODUCTION

Color is one of the most important quality attributes in food, because generally consumers get the impression that they like or dislike a food product based on its color (Andarwulan *et. al*., 2011). The process often decreases the color quality of food, therefore producers usually add synthetic colorant to improve the quality of the food (Wijaya & Mulyono, 2010).

Synthetic colorant tent to be chosen by manufacturers because of their high stability. However, the excessive use of synthetic colorant will have a harmful impact on health. With the development of the times, public awareness of health is increasing which makes the demand for natural food colorant as an alternative to synthetic colorant even higher.

One of the natural colorants commonly used in food comes from carotenoid compounds. Carotenoid compounds give food a yellow, orange to red color. Apart from being a dye, carotenoids have a role as a source of antioxidants and provitamin A which are beneficial to health (Amaya, 2016).

## STRUCTURE AND STABILITY OF CAROTENOID COMPOUNDS

Carotenoids are terpenoid compounds with color effects that range from red, orange, and yellow (Amaya, 2016). Violaxanthin, a xanthophyll, is a carotenoid member found in chloroplast membranes that causes a yellowish color. β-carotene and lutein from xanthophils are carotenoids found in the thylakoids of most plants (Janik *et. al*., 2008).

Carotenoids are tetraterpenoids (C40), which are a group of widely dispersed and fat-soluble pigments found in almost all plant species, from simple bacteria to yellow-flowered compositae. In plants, carotenoids consumed carotenoids, such as α-carotene and β-cryptoxanthin, also have provitamin A activity. Carotenoids are found in fruits and vegetables about 30-100% of human vitamin A requirements (Gross, 1991).

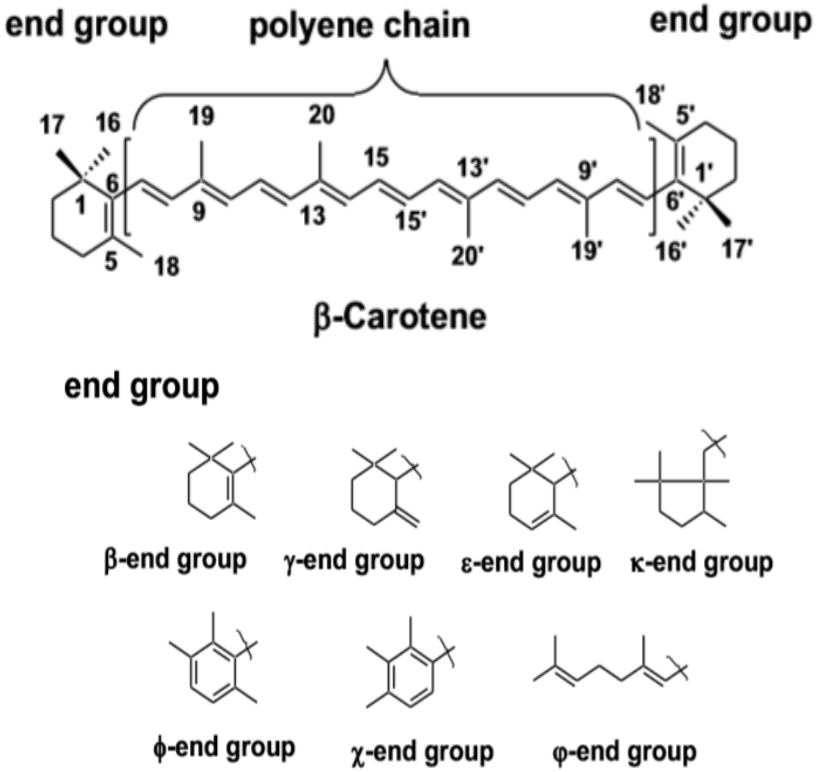


Figure 1. Basic structure of carotenoids and end groups (Britton *et. al*., 2004)

have two functions, namely as auxiliary pigments in photosynthesis and as colorant in flowers and fruits (Harborne, 1996).

Carotenoids are the most widespread pigments in nature. In higher plants, carotenoids in chloroplasts are often covered by the predominant chlorophyll pigment. In autumn when chloroplasts rot during plant aging, the yellow-orange color of carotenoids becomes clear (Fennema, 1996).

The role of carotenoid pigments is their ability as a precursor to vitamin A. Although β-carotene carotenoids have the greatest provitamin A activity due to their two β-ionone rings, other commonly

Most carotenoids consist of eight isoprene units with 40 carbon chains. The general structure of carotenoids generally consists of a polyene chain with nine conjugated double bonds and a final group at both ends of the polyene chain (Britton *et. al*., 2004). The chain structure of the polyene and the carotenoid end group is shown in Figure 1.

Carotenoids are divided into two groups, namely carotene and xanthophyll. Carotenes such as α-carotene, β-carotene, γ-carotene, and lycopene are hydrocarbons. Meanwhile, xanthophylls such as β-cryptosantin, lutein, zeasantin, astasantin, fukosantin, and peridinin, are carotenes containing oxygen atoms as hydroxy,

carbonyl, aldehyde, carboxylate, epoxide, and furanoxide groups in molecules (Maoka, 2009). The typical carotene and xanthophyll structures are shown in Figure 2.

stored in crystalline solid form and contain hydrocarbon solvents such as petroleum, hexane or benzene to minimize the risk of contamination with water before further analysis (Pinem, 2010).

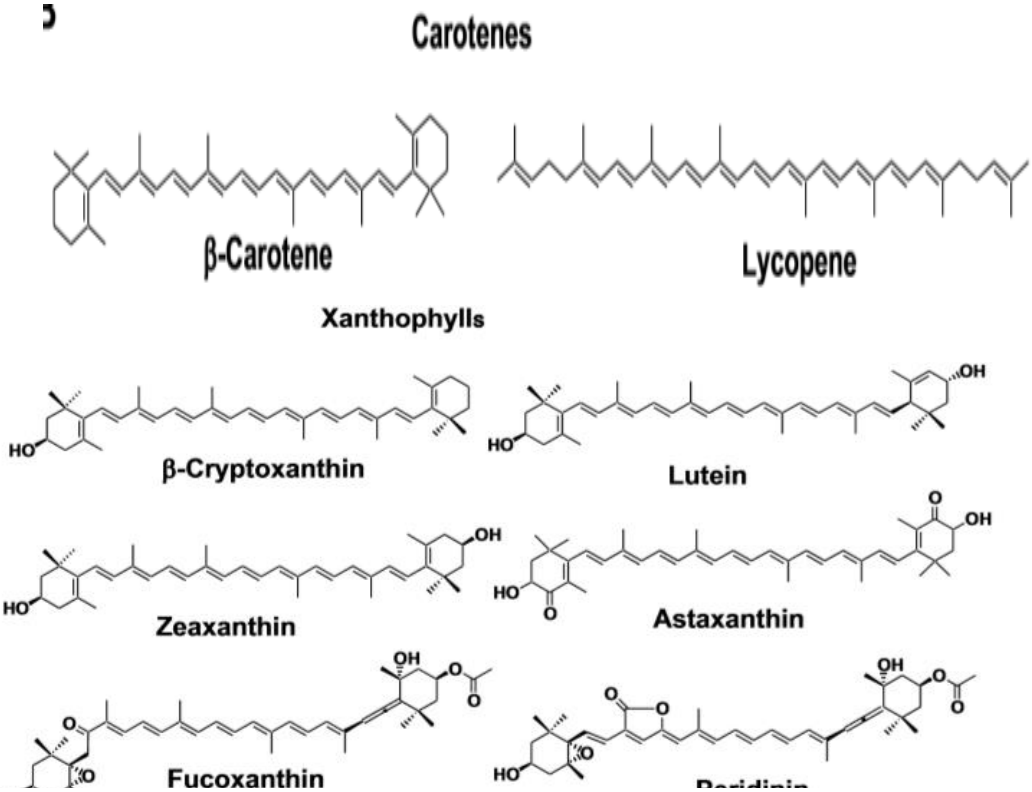


Figure 2. Structure of carotene and xanthophyll groups (Maoka, 2009)

The carotenoid most commonly found in plant tissue is β-carotene. These carotenoids are also used as food coloring. Several carotenoids found in plants, for example, are α-carotene found in carrots and capsanthin found in red peppers (Delgado-Vargas *et. al*., 2000).

All carotenoids are lipophilic compounds, thus carotenoids dissolve in oil and organic solvents, such as alcohol, chloroform, and acetone. Carotenoids are quite heat stable and their color can be lost due to the oxidation process (Thane & Reddy, 1997). In addition, carotenoids are very sensitive to acids, light, and oxygen (Friedrich, 1988), so they should always be stored in a dark room and in a vacuum, at a temperature of -200C. Carotenoids are best

Carotenoids are easily oxidized because of the many double bonds that are conjugated. In addition, the storage of carotenoid pigments in organic solvents will accelerate decomposition. This is due to the highly conjugated and unsaturated structure of carotenoids, so that the products of their degradation are very complex (Pinem, 2010).

During oxidation, epoxide and carbonyl compounds are initially formed. Further oxidation results in the formation of mono and oxygenated short chain compounds including epoxy-β-ionones. For provitamin A carotenoids, the formation of epoxides in the ring results in loss of provitamin activity. Extensive autoxidation will result in carotenoid pigment bleaching and loss of color. The oxidative breakdown

of β-carotene is intensified in the presence of sulfate and metal ions (Peisser & Yang, 1979).

Enzymatic activity, especially lipoxygenase will accelerate the oxidative degradation of carotenoid pigments. This process occurs through an indirect mechanism. Lipoxygenase will catalyze the oxidation of unsaturated or polyunsaturated fatty acids to produce peroxides, this is what causes lipoxygenase to react easily with carotenoid pigments (Ben *et. al*., 1971).

Carotenoids are relatively stable during storage and handling of most fruits and vegetables. The freezing process causes slight changes in carotene. However, the blanching process is known to affect carotenoid levels. Often the blanched plant products show a marked increase in the carotenoid content relative to the raw tissue. This is due to inactivation of lipoxygenase, which is known to indirectly catalyze oxidative decomposition of carotenoids, loss of water-soluble constituents or due to mild heat treatment which is usually used during the blanching process to increase the efficiency of pigment extraction (Francis, 1999). Although carotene is considered quite stable during heating, it is known that heat sterilization can induce a cis or trans isomerization reaction. To reduce excessive isomerization, this thermal process should be minimized whenever possible (Amaya, 2016).

Based on research conducted by Aryayustama *et. al*. (2018), showed that high storage temperatures could lead to a greater decrease in the total carotenoids of pandan fruit extract. In addition to storage temperature, the influence of the presence of oxygen can affect the structure of carotenoid compounds, this results in oxidation and isomerization of β-carotene pigments.

The color caused by carotenoids is from yellow to red so that the detection wavelength for monitoring carotenoids is usually in the range of 400-500 nm (Britton, 1995 in Susilowati, 2008). Hujaya (2008) reported that the maximum wavelength values of xanthophyll (444 nm) and carotene (450 nm) were not much different, but it was confirmed that the two compounds were different.

## CAROTENOIDS AS ANTIOXIDANT

Carotenoids are a group of pigments that can reduce free radicals, so they can act as antioxidants (Gross, 1991; Rodrigues-Amaya, 2003; Stahl & Sies, 2003). Therefore, carotenoids are able to protect cells and organisms from oxidative damage caused by free radicals. The buildup of free radicals will cause various health problems such as cancer, inflammation, Alzheimer's, cataracts, the aging process on the skin, and a decrease in the immune system. Free radical inhibition by carotenoids is mainly carried out by β-carotene (Limantara & Rahayu, 2012).

The role of carotenoids as antioxidants is as a reducer of singlet oxygen (1O2) and radical peroxides (Palozza and Krinsky, 1992; in Redriguez-Amaya, 2001; Miranda *et. al*., 1998). The way carotenoids work in reducing singlet oxygen is based on the transfer of electrons between the two molecules. Energy from singlet oxygen transfers to carotenoids, and then the oxygen ground state phase and triplet carotenoid excitation are obtained so that singlet oxygen reactivity can be reduced (Stahl & Sies, 2003).

The factor that really supports the function of carotenoids as antioxidants is their structure. The structure of carotenoids greatly affects their bioactivity, such as the presence of double bonds, open chains, and the least amount of oxygen substituents which will increase the antioxidant activity of carotenoids (Di Mascio *et. al*., 1989 in Lila, 2004; Dutta *et. al*., 2005; Tahamatsu *et. al*.,

2003). Following free radical stabilization by beta carotene can be seen in Figure 3.

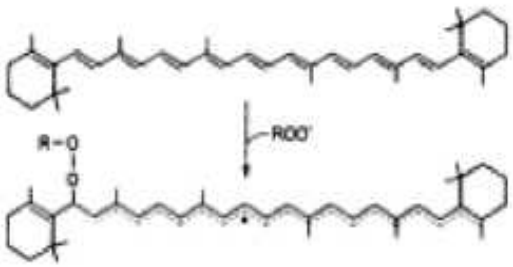


Figure 3. Free radical stabilization by beta-carotene (Belitz *et. al.*, 2004 in Nururrahmah *et. al*., 2013)

## SOURCES AND EXTRACTION METHODS OF CAROTENOIDS

The sources of carotenoids are very scattered in nature. The source can come from flowers, fruit, tubers, leaves, and so on. The following sources of carotenoids can be seen in Table 1.

Members of carotenoid compounds such as beta carotene, lycopene, and xanthophylls are also very scattered sources in nature. Following are the levels of beta carotene, lycopene, and xanthophylls can be seen in Table 2.

Table 1. Carotenoid contents from various sources

| Sources | Carotenoid Contents |
|---|---|
| Kabocha Pumpkin | 254.77 mg/100g |
| Yellow Pumpkin | 24.62 mg/100g |
| Seaweed (*Caulerpa* sp) | 12,532 mg/g |
| Cassava Leaves (*Manihot esculenta*, Crantz.) | 8,07 ± 45 µg/g wet basis |
| Rubber Cassava (*Manihot glaziovii*) | 767 ± 71 µg/g Wet basis |
| Pandanus Fruit Extract | 19,17%. |
| Yellow Ambon Banana Skin (*M. paradisiaca sapientum* L.) | 6,203 ± 0,004 µg/ |
| Sweet Potatoes with Yellow-Orange Tuber | 0,205-0,254 µg/100 g |
| Sweet Potatoes with White Tuber | 0,007-0,024 µg/100 g |
| Cilembu Sweet Potato | 1,363 ± 0,113 mg/g |
| Marigold Flower | 680 mg/kg |

Source: Manasika *et. al*. (2015); Kim *et. al*. (2005); Darmawati *et. al*. (2016); Magdalena *et. al.* (2007); Made *et. al*. (2018); Suparmi *et. al.* (2012); Qurniati *et. al*. (2013); Setyawati (2015); Yolanda (2012).

Carotenoids can be obtained by an extraction process using non-polar solvents or organic solvents. This is because carotenoids are intracellular and highly hydrophobic. The extraction methods that can be used to obtain carotenoids include the maceration extraction method, the supercritical fluid extraction method, and the enzymatic extraction method (Maleta *et. al*., 2018).

This maceration extraction method uses a solvent which diffuses into the cell of the material wherein the carotenoid compounds will come out as a result of osmotic pressure, besides that the maceration process is usually carried out by stirring and heating to speed up the extraction process. Solvents that are often used are acetone and ethanol (Maleta *et. al*., 2018).

Table 2. Beta Carotene, Lycopene, and Xanthophyll Content from Various Sources

| Type | Source | Content |
|---|---|---|
| Beta Caroten | Outer Dragon Fruit Skin | 181,6 ppm |
| | Inner Dragon Fruit Skin | 224,2 ppm |
| | Curly Red Chili (*Capsicum annuum* L Var. *Longum sendt*) | 5,57±0,13 mg/100g |
| | Cayenne pepper (*Capsicum frutescens* L.) | 0,36±0,01 mg/100g |
| | Big Red chilli (*Capsicum annuum* L. Var. *abreviatum Fingerhuth*) | 10,54±0,07 mg/100g |
| | Carrot Powder | 20550 µg/100 g |
| | Lompa Fish | 0,22 µg/g |
| | Purple Sweet Potato (*Ipomoea batatas*) | 75,91 ± 1,92 ppm |
| | Cantaloupe Fruit Extract | 3,171±0,150% |
| | Carrot | 34,94 ± 7,810 % |
| | Melon | 57,133 µg/g |
| Lycopene | Cilembu Sweet Potato | 0,038 mg/g |
| | Tomato | 3041 µg/110 gram |
| | Watermelon | 23,0 – 72,0 g/g |
| | Red Guava | 54 g/g |
| | Papaya | 20,0 – 53,0 gr/gr |
| | Red Grape | 33,6 gr/g |
| | Bali Orange | 1,38014±0,03007 mg/kg |
| | Forest Arben | 9 mg/100 g |
| Xanthophyll | Marigold Flower | 156,32 mg/kg |
| | Brown Seaweed (*Padina australis*) | 13,15 mg/10 g wet basis |

Source: Nururrahmah *et. al.* (2013); Octaviani *et. al.* (2014); Marliyati *et. al.* (2012); Mainassy *et. al*. (2011); Fauziah *et. al*. (2015); Kusbandari *et. al*. (2017); Agustina *et. al*. (2019); Idris (2011); Setyawati (2015); USDA National Nutrient Data Base (2020); Bramley (2000); Prihantini (2009); Tristiyanti *et. al.* (2013); Yolanda (2012) ; Nursid and Dedi (2017).

Things that must be considered in the extraction process using the maceration method are the extraction temperature and the stirring speed. The higher the temperature and the high stirring speed can accelerate the solvent to penetrate into the material and contact the material, but too high a temperature can also damage the bioactive components of the material (carotenoids) (Maleta *et. al*., 2018).

The supercritical fluid extraction method uses supercritical fluids which have the characteristics of low viscosity and relatively high diffusivity. One of the solvents often used in this method is liquid carbon dioxide because it has a critical temperature of 31.3 ℃ and a pressure of 72.9 atm. The main parameters that can affect the extraction with the supercritical liquid method are the ratio of the solvent to the material, the particle size of the material, the extraction temperature, the pressure, the extraction time, and the CO2 flow rate (Herero *et. al*., 2006).

The enzymatic extraction method uses the help of enzymes to extract the carotenoid compounds present in the material. The enzymes commonly used are

cellulase, pectinase, and hemicellulase enzymes. These enzymes will damage the cell walls of the material, so that carotenoid compounds can get out of the material. The factors that influence this method are pH and extraction temperature. The pH and temperature used are adjusted to the optimum conditions for the enzymes used (Lindahl *et. al*., 2013).

## CONCLUSION

Carotenoids are pigments that can be used as natural colorant in food. This pigment provides red, yellow, and orange colors. Carotenoids can act as antioxidants and provitamin A. Functions as antioxidants are caused by the carotenoid structure, which has double bonds, open chains, and the least amount of bound oxygen substituents. In addition, carotenoids are lipophilic compounds so they can dissolve in oil and organic solvents. Carotenoid pigments are quite resistant to heating, but they can be very easily degraded in acidic, light, and oxygen environments. The process of carotenoid pigment extraction can be done in three ways, namely maceration extraction, supercritical fluid extraction, and enzymatic extraction.

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