Review Article

# The Natural Pigments in Pigmented Rice Bran and Their Relation to Human Health: A Literature Review

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**Abstract**
Carotenoids and polyphenols are the natural pigments which are distributed to the pigmented rice bran. These pigments can prevent chronic disorders related to oxidative stress and antioxidant properties. This article focuses on carotenoids and polyphenols as ones among the natural pigments. Carotenoids and polyphenols exert an antioxidant property and anti-inflammatory agents; enhance immune responses and prevent Age-related Macular Degeneration (AMD). In combination with protein and dietary fiber, the bioavailability of carotenoids decreases in gastrointestinal tract. The inefficient absorption of anthocyanins in the small intestine resulted from unstability of the physiochemical pH and the degradation of anthocyanins in the small intestine. These have led to low bioavailability of anthocyanins.

**Keywords**: Pigmented rice bran, Carotenoids, Anthocyanins, Procyanidins, Absorption

## 1 Introduction

Traditional medicine in Asia has utilized various products from rice (*Oryza sativa* L.) for a long time. These uses of rice were known to the aboriginal people [1]. According to ethnopharma-cological studies, since ancient times, rice has been traditionally prescribed for therapeutic purposes, especially whole brown rice which is considered perfect food in various Asian countries including Thailand, Myanmar, China, Malaysia, Indonesia and India [1], [2]. For example, rice was used as medicine in China by royal Chinese physicians [1]. Malayan medicine stated that water, after soaking pounded raw rice or fine rice flour in it, could be used to treat eye diseases [3] and also to treat acute inflammation of inner body tissues. Dried

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rice powder has been applied to treat skin ailments. In Cambodia, the hull of mature rice plant was used to treat dysentery. The hulls of three-month-old rice plants were used as diuretic. In the Philippines, rice bran extract was used to prevent and to cure beri-beri as it is rich in vitamin B1 [4]. Moreover, in traditional Indian Ayurvedic medicine, rice is used to heal nutritional disorders, by consuming it as boiled rice with the starchy water and a small amount of salt [5]; this alleviates the pains in the digestive system. Furthermore, the same prescription also increased the virility of men.

Rice porridge administered orally can cure digestive disorders, such as constipation and gas pain, typhoid fever and hematemesis (vomiting of blood). The specific traditional rice Njavara has exhibited anti-inflammatory activity [6], [7]. Additionally, Chinese people claimed that rice as traditional medication could improve the spleen as well as the stomach, increase appetite and curing indigestion [1]. Hagiwara *et al*. [8] reported that pre-germinated brown rice contains small amounts of γ-aminobutyric acid (GABA), which may prevent patients with type II diabetes mellitus from macrovascular diseases associated with hyperglycemia and hyperlipidemia, such as ischemic heart disease, and can lower blood glucose concentrations. In addition, rice bran oil contains γ-oryzanol, which decreases the risks of obesity and of type II diabetes [9], reduces blood cholesterol both in *vivo* and in a clinical study [10], replaces chemotherpy [11], [12], antibacterial [13], and stimulates hair growth and prevents skin aging [14].

Pigmented rice is considered as a functional food in China, Japan, Korea and India due to its high polyphenol and anthocyanin contents [16]. However, the nutrient composition of rice depends on its cultivation [17], and its contents of bioactive compounds also depend on genetic characteristics of the rice variety [18], [19]. Some cultivars of rice have pigments on the pericarp and seed coats, causing red, black or purple coloration. When comparing pigmented and white rice brans nutritionally, the pigments act as sources of phytochemicals. These include hydrophilic and semi-hydrophilic compounds (anthocyanins and polyphenols) which usually disseminate in the pericarp and aleurone layers of the grains [20]–[22]. In addition, black rice bran is rich in lipophilic compounds including carotenoids [23],

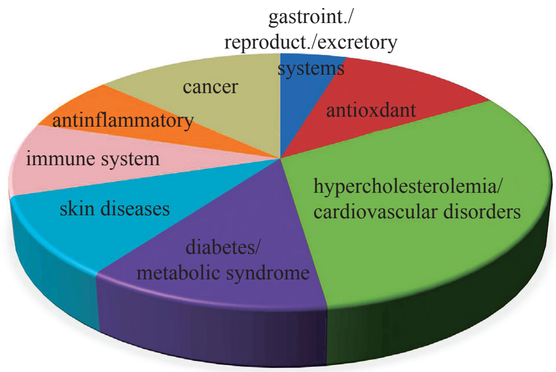


**Figure 1**: Therapeutic functions of rice and pigmented rice [15].

[24], tocopherols, tocotrienols and γ-oryzanol, which are distributed in the outermost pericarp, germ and aleurone layers [25], [26]. All these compounds may provide health benefits and prevent chronic disorders related to oxidative stress and antioxidant activity [27]–[29]. The therapeutic roles of rice-derived products and their constituents were curated from literature as shown in Figure 1. This review focuses on the naturally occurring pigments in pigmented rice bran, such as carotenoids and anthocyanins.

## 2 Carotenoids in Cereal Grains

Carotenoid pigments in nature provide red, yellow or orange coloration to fruits and vegetables. These compounds are also found in plant seeds [30]. They include $\alpha$- and $\beta$-carotene, lutein, lycopene and zeaxanthin, which are red and yellow in colour (Figure 2). These are distributed in the outermost pericarp, germ and aleurone layers of black rice bran [23], [24]. Some studies have reported that the predominant carotenoids in wheat are lutein and two non-pro-Vitamin A xanthophylls, which are oxygenated derivatives of carotenes [31], [32]. Their chemical structures include hydroxyl groups, ones on each side of the molecule. Xanthophylls are more polar than carotenes and tend to be distributed between LDL and HDL [33]. Their hydroxyl groups may play an important role in the antioxidant function.

Carotenoids, which act as radical scavengers and singlet oxygen quenchers [34], decrease the risk of degenerative diseases, such as cancer, cardiovascular diseases, and age-related macular degeneration, and

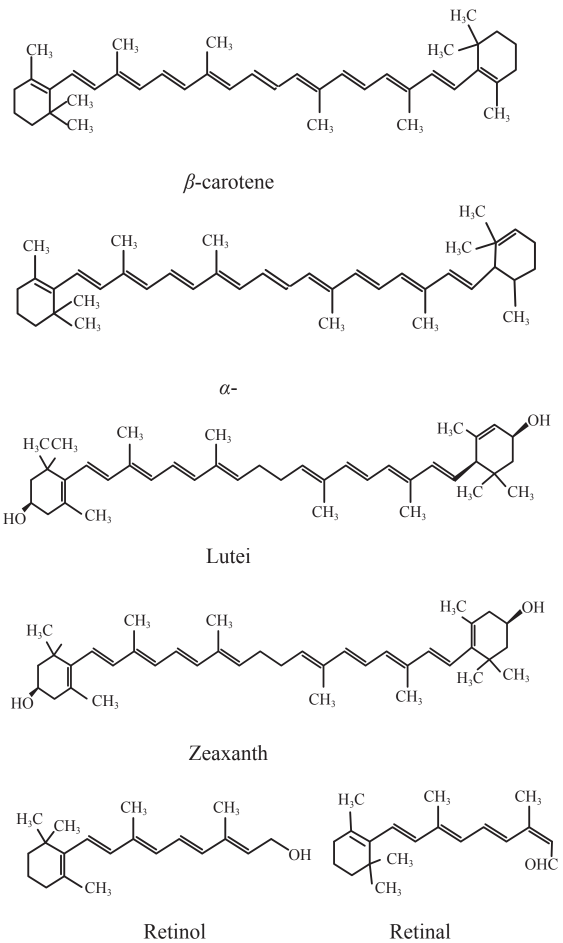


**Figure 2**: Chemical structures of the carotenoids in cereal grains.

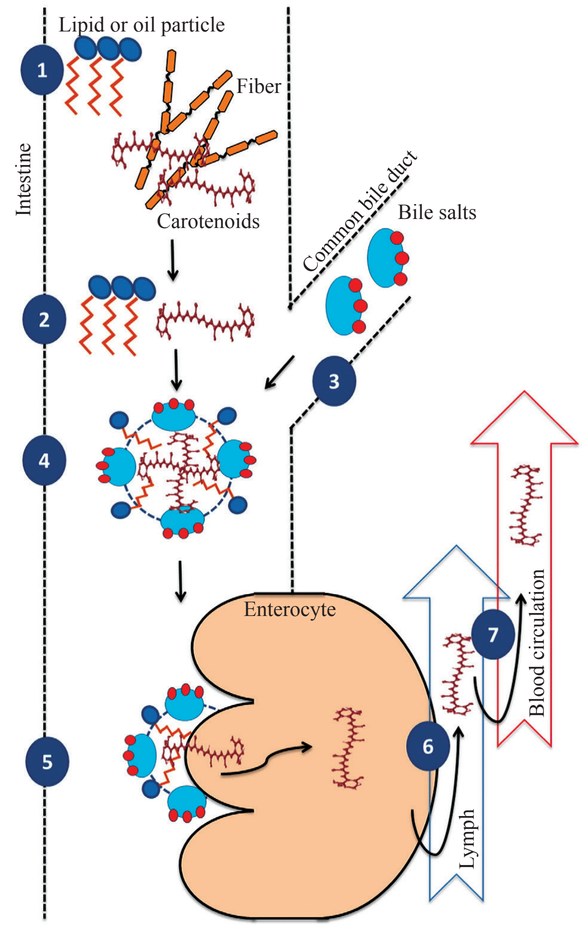


**Figure 3**: The absorption of carotenoids from the food matrix. 1) and 2) Disruption of food matrix, 3) Release of bile salts from the common bile duct, 4) Uptake of carotenoid molecules by a lipid droplet and formation of a micelle, 5) Uptake of carotenoid molecules in enterocyte, and 6) Release of carotenoid molecules to blood circulation [79].

help maintain skin health [35], [36]. They also act as an anti-inflammatory agents, enhance immune responses [37] and prevent Age-related Macular Degeneration (AMD) [33]. In 2007, the World Cancer Research Fund (WCRF) reported that carotenoid intake and breast cancer are associated; therefore, high *α*-carotene, *β*-cryptoxanthin, lutein, (+)zeaxanthin, lycopene and total carotenoids levels in bloodstream can decrease the risk of breast cancer [38]. Approximately, 40 carotenoids are present in the human diet, 20 carotenoids in human blood and tissues [37], while 6 carotenoids are found in human serum, namely *β*-cryptoxanthin, lutein, lycopene, zeaxanthin, *β*-carotene and *β*-carotene [33]. *β*-carotene and 50 of the carotenoids can be metabolized and converted to vitamin A. Moreover, among these carotenoids, *β*-carotene has the highest provitamin A activity [33].

When carotenoids enter the digestive system, they are incorporated into micelles in the gastrointestinal tract and absorbed by mucosal cells of the small intestine [39]. Primarily, carotenoids are partly cleaved and converted to retinylester. Carotenoids and retinylester are bound to chylomicrons before entering the lymphatic system and the blood circulation, respectively, as shown in Figure 3 [40].

Because of the complexation of carotenoids with soluble protein and dietary fiber, they are captured in the plant cell walls, the bioavailability of $\beta$-carotene becomes quite low (10–65%) [41]. When complexing with protein, this inhibits its transportation into the gastric emulsion with bile salts. Thus, the gastric emulsion will always inhibit the absorption of carotenoids in the small intestine. The release of carotenoids from the food matrix depends on the degree of digestion, which may be assisted by mechanical processing reducing the particle sizes prior to digestion. Increase in specific surface would help the contact with pancreatic lipases and bile salts, improving digestion and release [42]. The absorption of carotenoids would begin at the micelle formation because the micelles always affect their bioavailability. Additional dietary fat could also improve the bioavailability of carotenoids [43]. The addition of extra oil usually benefits non-polar carotenoids more than polar carotenoids (xanthophylls) [44]. Also, other factors, such as thermal processing and structural barriers in food (matrix, cell wall integrity, bio-encapsulation), have influenced on bioaccessibility. The lipids which can be added to improve the bioavailability are the most important factor determining the bioavailability of carotenoids.

## 3 Polyphenol in Cereal Grains

The phenolic compounds present in rice include phenolic acids, flavonols, anthocyanins, and procyanidins. Generally, non-pigmented rice varieties provide only phenolic acids, whereas pigmented rice is richer in polyphenol compounds, especially in anthocyanins and procyanidins. For instance, red rice contains procyanidins, while black rice contains either anthocyanins or procyanidins, or both, depending on the variety [45]. Phenolic acid exists in free, soluble conjugated, and insoluble bound forms. All of these are highly concentrated in the bran fraction [46], [47]. There are 6 types of phenolic substances in raw black waxy rice bran, namely gallic acid, (+)-catechin, p-coumaric acid, syringic acid, ferulic acid and caffeic acid. Gallic acid is the major phenolic acid, followed by ferulic acid, while syringic acid contributes the smallest amount. These phenolic substances play an important role in the antioxidant activities and offer health benefits for those who suffer from the chronic disorders [48]–[51]. Some reports have indicated that the total phenolic content in white rice or unpolished rice (14.6–33.4 mg/100 g) and red rice (66.8–422.2 mg/100 g) was higher at 1 week of development in the flowering stage than in the mature stage, whereas black rice had higher antioxidant activity at maturity [52], [53]. Rice bran showed the largest total phenolic content, especially as bound fraction. From Table 1, total phenolic contents of red and black pigmented rice were compared with the white rice grains. The phenolic acids in white, red and black rice bran accounted for 88%, 89% and 91%, respectively. White rice had significantly lower levels of phenolic acids than pigmented rice bran, whereas black rice bran showed significantly higher levels of total phenolic contents than red rice bran. Protocatechuic Acid (PA) content was detected only in black and red rice bran. Regarding $p$-Hydroxybenzoic Acid ($p$-HA), it was detected in red and white rice bran. The Syringic Acid (SRA) content in red and white rice bran was significantly higher than that in black rice bran. Trans-p-coumaric (trans-$p$-CA), Ferulic Acid (FA), trans-sinapic acid (trans-SNA), and Isoferulic Acid (IFA) contents in white rice bran were higher than those in red and black rice bran.

**Table 1**: Total phenolic contents of white, red, and black rice grains [53]

| | Free[b] | Free/ Conjugated[b] | Bound[b] |
|---|---|---|---|
| **Whole Grain** | | | |
| White[c] | 0.07±0.01 | 0.14±0.02 | 0.19±0.06 |
| Red[c] | 0.17±0.00 | 0.70±0.003 | 0.50±0.04 |
| Black[c] | 0.25±0.001 | 0.42±0.01 | 0.63±0.02 |
| **Embryo** | | | |
| White[c] | 0.42±0.05 | 1.46±0.09 | 0.94±0.09 |
| Red[c] | 0.44±0.01 | 1.49±0.03 | 1.28±0.07 |
| Black[c] | 0.53±0.04 | 1.80±0.08 | 1.40±0.17 |
| **Endosperm** | | | |
| White[c] | 0.05±0.00 | 0.05±0.001 | 0.03±0.00 |
| Red[c] | 0.05±0.00 | 0.05±0.001 | 0.08±0.02 |
| Black[c] | 0.06±0.01 | 0.07±0.001 | 0.05±0.03 |
| **Bran** | | | |
| White[c] | 0.30±0.04 | 1.08±0.03 | 3.93±0.27 |
| Red[c] | 1.66±0.03 | 6.42±0.35 | 3.74±0.41 |
| Black[c] | 1.53±0.02 | 3.09±0.04 | 3.78±0.01 |

The results are presented as mg gallic acid equivalent/g rice sample.
[b] Values in each column with different capital letters are significantly different ($p < 0.05$).
[c] Values in each row with different lowercase letters are significantly different ($p < 0.05$).

**Table 2**: Total anthocyanin content and anthocyanin composition of black rice grain [53]

| | TAC[a] (mg cyanidin -3-glucoside equivalent/g) | Cyanidin 3-O- glucoside[a] (mg/g) | Peonidin 3-O- glucoside[a] (mg cyanidin 3-O-glucoside equivalent/g) | Cyanidin 3-O-rutinoside[a] (mg cyanidin 3-O-glucoside equivalent/g) |
|---|---|---|---|---|
| Whole grain | 0.87±0.02 | 0.52±0.02 | 0.14±0.01 | 0.06±0.01 |
| Embryo | 0.34±0.01 | 0.12±0.01 | 0.09±0.01 | nd |
| Endosperm | nd | nd | nd | nd |
| Bran | 6.28±0.10 | 3.58±0.002 | 0.70±0.003 | 0.28±0.01 |

TAC = Total Anthocyanin Contents

Anthocyanins are a group of hydrophilic flavonoids. They are natural colorants responsible for red, purple and black colors of pigmented rice. The major anthocyanin in purple bran is cyanidin-3-glucoside [20]. Types of anthocyanins in black rice included cyaniding 3-O-glucoside (C3G), peonidin 3-O-glucoside (P3G) and cyanidin 3-O-rutinoside (C3R). The fractions of total anthocyanin in bran and in embryo of black rice were 97% (6.28±0.10 mg C3G equiv/g) and 3% (0.34±0.010 mg C3G equiv/g), respectively. From Table 2, C3G and P3G in black rice were detected only in bran and embryo, whereas C3R was found in black rice bran [53].

When the effective concentrations of anthocyanins were compared both in *vitro* and in a clinical study, in the latter plasma concentrations were lower than those in *vitro* [54]–[56]. The absorption of anthocyanins in the small intestine was poor; only about 1% of the total amount was administered orally. When being exposed to the physiochemical pH of the small intestine, the anthocyanins are unstable and are degraded to other metabolites, which reduces their bioavailability [57]. Regarding absorption, phase II metabolism takes place in the gut or liver [58], [59]. The phenolic acids (i.e. protocatechuic acid) are degraded spontaneously or by microbial fermentation in the colon [60].

Antioxidant properties of the bioactive ingredients in plant-derived foods have been widely studied. In experiments simulating gastric digestion in *vitro*, the total anthocyanin content and the scavenging capacities of DPPH and ABTS radicals were not significantly affected by those of undigested sample, whereas the purple rice anthocyanins were remarkably reduced by the simulated intestinal digestion [61]–[63]. Anthocyanins could reduce exposure to ROS which usually contributes to the incidence of chronic diseases. An $H_2O_2$ induced cell injury model is usually used to assess cytoprotective effects. Regarding such

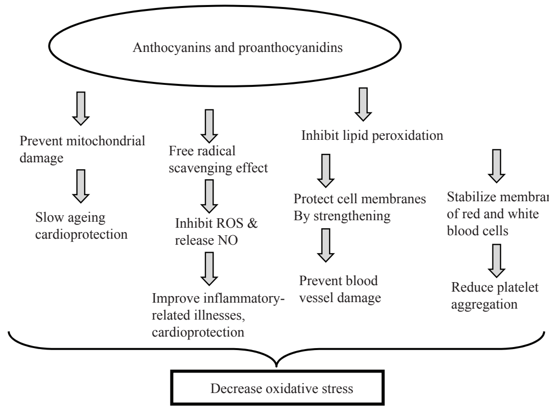


**Figure 4**: A decrease in oxidative stress by antioxidant such as anthocyanins and proanthocyanidins and free radical scavenging effects. ROS = reactive oxygen species, NO = nitric oxide [80].

cytoprotective activity, cyanidin-3-glucoside could increase the levels of Superoxide Dismutase (SOD) and Glutathione (GSH) more than peonidin-3-glucoside [64], conferring prevention of renal oxidative stress in animal experiments [65]. When considering anti-inflammatory effects, cyanidin-3-$\beta$-D-glycoside from black rice and its metabolites, cyanidin and protocatechuic acid have high reducing power [66] and can inhibit nitric oxide [67]. They also can release TNF-α, IL-1α, NO, and PGE2, induce activation of NO synthase (NOS) and COX-2 genes, and activate NF-κB and MAP kinases [68]. Proanthocyanidins from red rice and anthocyanins (cyaniding 3-O-glucoside and peonidin 3-O-glucoside) in black rice have decreased the concentration of nitric acid, as well as of other Reactive Oxygen Species (ROS), both *in vitro* and *in vivo*. In *in vivo* studies, they also increased the antioxidant capacity and activity of antioxidant enzymes, such as Superoxide Dismutase (SOD) and Catalase (CAT) [69]. Furthermore, black rice anthocyanidins can protect the retina against photochemical damage and inhibit apoptosis of retinal cells, which is otherwise induced by fluorescent light in an animal model [70]. In relation to cardiovascular prevention, cyanidin-3-glucoside from black rice anthocyanin improves both hypercholesterolemia and hyperlipidaemia, and simultaneously decreases hepatic lipogenic enzymes in hyperlipidaemic rats [71]. In summary, anthocyanins and proanthocyanidins have noticeable ability to prevent or reduce oxidative stress, as shown in Figure 4.

(+)Catechin and epicatechin are the monomer units of proanthocyanidins, which are polymers of flavonol. Proanthocyanidins can shield against cancer and cardiovascular disorder [72], [73]. Regarding bioavailability, the trimers of proanthocyanidins may be absorbed in the gastrointestinal tract [74]. Proanthocyanidin may be degraded into oligomers with DP $< 3$, mixed with epicatechin monomer and dimers in the acidic stomach environment [74]. Finally, it is absorbed in the small intestine [74], [75]. The proanthocyanidins with DP $> 10$ are degraded in the cecum large intestine by colonic microbiota [76]. The intestinal absorption of proanthocyanidins has positive impacts on health [77]. However, the mechanism of absorption and bioavailability of proanthocyanidins is still under continuing studies.

**Table 3**: Total proanthocyanidin contents in whole grain rice with red rice bran [68]

| Proantho-cyanidin | Concentration (µg/g grain) | Contribution (%) |
|---|---|---|
| Monomers | 5.30±0.33 | 0.42 |
| Dimers | 25.48±0.37 | 2.00 |
| Trimers | 52.45±0.75 | 4.12 |
| Tetramers | 87.50±1.18 | 6.87 |
| Pentamers | 123.69±1.91 | 9.72 |
| Hexamers | 215.56±5.89 | 16.93 |
| Heptamers | 128.22±1.67 | 10.07 |
| Octamers | 121.99±1.87 | 9.58 |
| Nonamers | 81.69±2.34 | 6.42 |
| Decamers | 56.90±4.92 | 4.47 |
| Polymers | 374.10±6.67 | 29.39 |
| Total | 1272.89±22.69 | 100.00 |

From the data in Table 3, 5–8 mer oligomers accounted for 40% of proanthocyanidins in red rice bran, while oligomers with more than 10 mers accounted for 29%. In Japanese red rice bran, the total proanthocyanidins contents accounted for 71% which higher oligomers for 4–6 mers and lower oligomers for 1–3 mers. The bioactivity of proanthocyanidins depends on their structure. For example, the procyanidin fractions with DP $< 5$ and DP $> 5$ the mechanisms of homeostatic modulation of immune function and inflammation are different [78].

## 4 Conclusions

Due to the increasing costs of medical treatment, an alternative strategy is to maintain good health by preventing risks of degenerative diseases. Phytochemicals from plants can contribute to this strategy. Rice is the main staple food globally. In Ayurvedic medicine, white rice has contributed to therapeutic applications, but it is pigmented rice that is rich in phytochemicals. The natural pigments in pigmented rice bran include carotenoids, anthocyanins and proanthocyanidins. These can decrease the risks of degenerative human diseases. Carotenoids which usually act as anti-inflammatory agents can improve the immune response. Polyphenols, such as anthocyanins and proanthocyanidins, exert antioxidant and anti-inflammatory activities, and they can treat hypercholesterolemia and hyperlipidaemia. There is considerable evidence motivating studies of dietary pigmented rice in terms of its positive health effects. Supplementing human diet with pigment extracts from pigmented rice bran has not yet been much investigated, but it might be relevant to cardiovascular or cardiomyopathy mortality from cardiovascular diseases.

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The authors have no conflicts of interest to declare.

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