Pigmented Rice: A Quality Food for Human Nutrition

T. B. Bagchi,[1*] K. Chattopadhyay,[1] A. Kumar,[1] P. Sanghamitra,[1] S. Sarkar,[1] and M. Sivashankari [1]

[1]ICAR-National Rice Research Institute, Cuttack, Odisha, India.

*Corresponding Author:

T. B. Bagchi

Email:
torit.crijaf09@gmail.com

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SUMMARY

Pigmented rice had been considered as a super food since Chinese emperor's era due to its higher nutritive value as compared to white rice. Unfortunately, it is not so popular throughout the world for its unacceptable physiological traits and inferior physical quality of the grain. But it is rich in many bioactive molecules, which is directly associated with health benefits of human beings. The present review paper mainly focuses on biochemistry and molecular biology of nutritional compounds as well as sensory studies of pigmented rice. It also revealed the genetic approach for the improvement of the grain qualities. Pigmented rice is rich in antioxidants like phenolics, oryzanol, vitamins and minerals in some cases. They are poor in head rice recovery and generally low amylose content. Due to lower amylose content, the grains are waxy in nature and therefore, it can be utilised for the preparation of many rice based products.

INTRODUCTION

Functional foods are defined as the foods that are consumed as a part of usual diet with normal quantity and having good sensory quality that promote health through enhanced immune response and minimizing disease infection so that the aging process become slow down. These types of food may be especially processed or unprocessed plant and animal products that contain higher concentration of bio-active compounds in addition to normal nutrients, present in conventional foods. Pigmented rice particularly black rice is considered as a functional food in Indonesia .Red rice is also recognised as a functional food in Japan because of its high phenolics and anthocyanin content. In India, China and other South East Asian countries, black rice is utilised to promote health and combat many diseases. There are various types of pigmented rice exist in the world such as red, purple, yellow and black. Interestingly, pigmentation of rice is mainly associated with bran or pericarp layers not with the edible endosperm of the grain. As brown rice is not so popular among the consumers, the percent adherence of coloured bran with the endosperm after milling is very important to get benefits of pigmented rice. Pigmented rice is enriched with many bio-active compounds and these bio-active compounds like anthocyanin, phenolic acids, flavonoids, pro-anthocyanidins, tocopherols, tocotrienols, γ-oryzanol are present in higher quantity as compared to white rice. Therefore, it exhibits higher rate of antioxidant activity also. These antioxidant compounds scavenge reactive oxygen species (ROS) such as lipid peroxide and superoxide anion radicals and lower cholesterol content of animals.

Black rice is a good source of fibre and minerals besides basic nutrients. Anthocyaninis the prime component of pigmentation in case of all the pigmented rice cultivars. Pigmented rice is rich in medicinal properties. The red rice cultivar of South India Njavara was used to treat arthritis , cervical spondylitis, muscle wasting, skin diseases and certain neurological problems. Previous investigators demonstrated that black rice diet significantly inhibits atherosclerotic plaque formation in rabbits.

Chemistry behind pigment formation in rice

The red grain colour is due to the deposition and oxidative polymerization of pro-anthocyanidins in the pericarp, whereas the black grain colour is caused by deposition of anthocyanins. Anthocyanin is sequestered in the vacuole through ABC type transporter and plays the defensive role in the plants. In many instances, it is the inherent colour of the grain and plant parts. It is synthesised from phenylpropanoid acetate pathway and the precursors are three molecules malonyl-CoA and one molecule 4-coumaroyl-CoA. The main regulatory enzyme is Chalcone synthase, which facilitate the first committed step of anthocyanin synthesis to form Naringenin Chalcone (Figure-1). However, 4-coumaroyl-CoA is synthesised from an amino acid L-Phe (synthesizes from Shikimate pathway) with the help of Phenylalanine ammonia lyase (PAL), as a regulatory enzyme.

Flavonoids consist of 15-carbon skeleton that is organized in two aromatic rings (A- and B-rings) interlinked by a three-carbon chain (structure C^1-C_3-C6).The most basic flavonoids the flavones, flavonols, and flavanones are widespread in the plant kingdom and typically absorb damaging UV rays. Proanthocyanidins (PAs), also known as condensed tannins, are a major subgroup of flavonoids that are oligomers and polymers of flavan-3-ol units. Flavonoids are recognized for both their ability to donate electrons and to stop chain reactions, which produce free radical. These antioxidant activities are attributed to the phenolic hydroxyls, particularly in the 3OH and 4OH of the three-carbon chain. Seven flavonoids are reported in rice of which tricin appears to be the major flavonoid in the bran. Other flavonoids present are decreased in the following order: luteolin (14%)

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>apigenin(6%) >quercetin (3%) >isorhamnetin (1%) >kaempferol (<1%) >myricetin (<1%).The biosynthetic genes involved in flavonoid synthesis are hardly expressed in endosperm (Rice Expression Profile Database (Rice Xpro), http:// ricexpro. dna.affrc.go.jp/pro/), while in embryos the enzymes Phenylalanine ammonia lyase (PAL), Chalconeisomerase (CHI), Chalcone synthase (CHS), and Flavone synthase (FNS), which are thought to be involved in tricin biosynthesis, are expressed .Earlier workers reported that the red and black rice grains had higher flavonoid content than non-pigmented rice. It was also noticed that all the flavonoids found in rice leaves are flavones (luteolin, apigenin, tricetin etc.) derivatives.

Genetic analysis of grain antioxidants and pigment formation

The understanding of genetic basis for the formation of phenolics, flavonoids, and antioxidant capacity is very important with respect to pigmented rice. It was observed through linkage mapping that the synthesis of these constituents were seperately controlled by three QTLs. Among those QTLs only one QTLs is present on chromosome 2 and it is responsible for phenolic content and flavonoid content of grain. Through association mapping, The previous researcher also identified the QTLs for these traits utilizing a diverse set of red rice and black rice germplasms., Four QTLs for phenolics , six QTLs for flavonoids and six QTLs for antioxidant capacity were identified. Among them, four QTLs for phenolic content were also shared for other two traits. Rc (brown pericarp and seed coat) and Ra (Prp-b for purple pericarp) were main-effect loci for grain color and nutritional quality traits. After association mapping of the 361 white rice germplasms, it was found that marker RM346 is related with phenolic content. The black rice generally accumulates anthocyanins whereas red rice synthesize pro-anthocyanidin which acts as antioxidants for human beings. The bran color of red rice is generally controlled by two complementary genes, Rc on chromosome 7(brown pericarp) and Rd on chromosome 1(red pericarp). Red seed color is produced when these loci are present together. Rd alone has no phenotype but Rc in the absence of Rd produces brown seeds. A natural mutation in rc has reverted brown bran to red bran and resulted in a new dominant wild-type allele, Rc-g. The dark purple bran colour of rice was controlled by two complementary genes, Pb (on chromosome 4) and Pp (on chromosome 1). After mapping of Pb gene, the researchers observed that this gene may be Ra gene. Therefore, the markers for these genes may be useful for pigmented rice breeding.

Sensory and yield parameters of pigmented rice

Sensory qualities of rice grain and different products are important factors for consumer acceptance. It includes texture profile of the grain and products, aroma, colour, physicochemical and cooking properties, viscosity of rice flour and physical impurities. Textural profile analysis of rice grain and its various parameters includes hardness, fracturability, adhesiveness, cohesiveness, chewiness, gumminess and springiness. The cooking methods significantly affect the textural properties of cooked rice while the effect depends on the rice genotypes and method of processing. Previous worker pointed out that amylose content of grain determines the hardness of cooked rice. The rice grains with higher amylose content are normally less sticky after cooking and hard on cooling. It was reported that hardness arises from different arrangement of starch granules in different products, as evident from SEM micrograph.

In rice, aroma is one of the major sensory quality for consumer preference. The odor-active volatile compounds (2-acetyl-1-pyrroline, guaiacol, hexanal, isovaleric acid, Valeric acid, enantic acid, 2-methylnaphthalene, 2-nonenal, octanal, mrystic acid, 1-octen-3-ol,2-heptanone, heptanal,) contribute the most in different pigmented rice types. In black aromatic rice 2-acetyl-1-pyrroline is the most abundant compoud.

The grain yield of cereals is determined by the balance between the sink size and the source capacity and the reduced sink size of black rice is responsible for reduced yield. The farmer's preference to opt for a variety for cultivation depends partly on the duration of the crop and variety with short duration and good yield is always preferable to farmers. The major constraint with red pigmented rice is its low head rice recovery (HRR) than white rice. Black rice cultivars are more suitable to promote health improvement owing to its fused bran layer with endosperm, which retains the nutritional compounds and render antioxidant capacity of milled grains. Amylose is the main biomolecule that determines texture of cooked rice i.e. stickiness or looseness of the grain after cooking. Higher amylose content (>20%) is generally associated with looseness of cooked rice and vice-versa and amylose content of the pigmented rice grains is shown to be affected by environmental factors.

Nutritional advantages of pigmented rice cultivars over white rice

(a) Phenolic Acids and Flavonoids

Prevoius study reported that the pigmented rice samples had higher content of phenolic compounds than non-pigmented samples and the pericarp layers (bran)was the major source of various phenolic acids (ferulicacid, *p*-coumaric acid, vanillic acid, *p*-hydroxybenzoic, gallic acid, and protocatechuic acid). In addition, the brown rice was also rich in different phenolic compounds (0.05 to 7.69 mg/100g) than the polished rice (0.02 to 1.12mg/100g) with the predominance of p-coumaric acid, ferulic acid, gallic acid and protocatechuic acid.The free phenolics content in black rice bran ranged from 2086 to 7043 mg of GAE/100 g (DW) constituting 88.2 to 95.6% of total acids. The bound phenolic content ranged from 221.2 to 382.7 mg of gallic acid equiv/100 g (DW). In fact, Phenolic compounds exist in three forms:

TABLE 1. Some Inorganic Ions That Serve as Cofactors for Enzymes.

Sl.No.	Ions	Enzymes
1	Cu^{2+}	Cytochrome oxidase
2	Fe^{2+},Fe^{3+}	Cytochrome oxidase, catalase, Peroxidise
3	K^{+}	Pyruvate kinase
4	Mg^{2+}	Hexokinase, glucose 6-phosphatase, pyruvate kinase
5	Mn^{2+}	Arginase, ribonucleotidereductase
6	Mo	Dinitrogenase
7	Zn^{2+}	Carbonic anhydrase, alcohol dehydrogenase, carboxypeptidases A and B

the free, soluble conjugate (esterified), and insoluble-bound forms. Due to presence of carboxylic acid and hydroxyl groups in their structures, phenolic acids form both ester and ether bonds with other compounds resulting the formation of linkages with cell wall polysaccharides. Phenolic acids can be esterified with small molecules like alcohols, other phenolic acids and alkaloids to form soluble conjugated structure. However, it was reported that there was 20 to 26 % reduction of total phenolics content and 13 to 20 % reduction in total flavonoids content in some black rice during 25 minutes domestic cooking. It was also reported that diet containing black rice reduced the level of plasma cholesterol, triglycerides, and low-density lipoprotein and increase the level of good lipid (HDL) in rats.

FIGURE 1. Unmilled grain of some red and black rice.

(b) Gamma-Oryzanol Content

Chemically, γ-oryzanol is the esters of ferulic acid with phytosterols and triterpene alcohols, generally referred as steryl ferulates. Acetyl-CoA is the precursor molecule for biosynthesis of γ-oryzanol and form isopentenyl diphosphate (IPP) via mevalonic acid through some enzymatic steps. Previous study reported that γ-oryzanol content of 196 Korean landraces ranged from 9.8 to 55.9 mg 100 g^{-1} and a mean was 27.2 mg /100 g. However, cooking generally decrease the γ-oryzanol content of the grain with a few exception. The previous researchers pointed out that genotype and environment factors had a significant role in controlling gamma oryzanol level per grain weight. γ- Oryzanol content of pigmented rice was higher (44.73mg/ml rice extract) than non pigmented (0.0 to 2.5 mg/ml rice extract) genotypes. The concentration of γ-oryzanol varied in different components of the grain. The highest concentration of it was found in the rice bran, obtained from the first 10 s milling. In other words, it can be said that the highest γ-oryzanol concentration was found in the outer portion of the bran layer. γ-Oryzanol plays a significant role in various biological activities, including anti-inflammatory , antioxidant, and anti-tumor activities. In addition, γ-oryzanol has also been reported to lower blood cholesterol levels in humans .

(c) Vitamin-E Content

Vitamin E was discovered by Even and Bishop. Vitamin E is necessary for human reproduction and it prevents reactive oxygen linked diseases viz. cardiovascular disease, cancer, chronic inflammation and neurologic disorders. Vitamin E deficiency leads to irreversible disorder like muscle weakness, peripheral neuropathy and ophthalmoplegia. Vitamin E is absorbed into intestinal mucosa by passive diffusion process. Human body can store 40 mg/kg vitamin E, 77% of which is in adipose tissue. At transcriptional level, several genes like tocopherol transfer protein [α-TTP], α-topomyosin and collagenase etc. are controlled by α-tocopherol. In rice, vitamin E homologs are varied according to the genotypes and post harvest processing. In Brazil, thirty-two different samples of dehulled brown rice (Indica and japonica) were analysed for tocopherol and tocotrianol content and observed that total vitamin E levels varied largely at 10.4–32.5 mg/kg. In, *japonica* subspecies, the average concentrations of different tocols from high to low were α-Tocopherol (10.0 mg/kg), α-Tocotrianol (7.0 mg/kg), γ- Tocotrianol (5.8 mg/kg), and γ- Tocopherol (1.4 mg/kg). In indica rice, the most abundant homolog was γ- Tocotrianol (7.8 mg/kg), followed by α-Tocopherol (4.8 mg/kg), then α- Tocotrianol (2.3 mg/kg), and γ-Tocopherol (1.3 mg/kg). It was found that vitamin E levels were greatly reduced after parboiling of rice grain.

(d) Minerals Content

Micronutrients (Zn, Mn, Cu and Fe) are very essential elements in our body where they acts as a regulator of metabolic functions, fluid balance, blood pressure, nervous and immune system. Minerals also acts as a cofactor of different enzymes in our body. K and Mg were the most abundant minerals in whole rice grains, accounting for about 60%and 30% of total minerals, respectively. Mn and Zn showed higher content in black rice than in most of non-pigmented and red rice samples.

Among all the minerals, phosphorus content (2062.1 to 2529.7 mg/kg) in pigmented brown rice (Chak-hao Angangba, brown rice; Chak-hao Poireiton, purple rice and Chak-hao Amubi, black rice), was the highest, followed by potassium (1546.8 to 1843.6 mg/kg), sulphur (743.5 to 976.1 mg/kg), magnesium (377.2 to 387.6 mg/kg), calcium (77.6 to 136.2 mg/kg), iron (47.2 to 88.8 mg/kg), and zinc (34.9 to 53.9 mg/kg), while copper content (27.5 to 33.4 mg/kg) was the lowest. However, polishing of rice led to significant declines (0.75 to 2.50 times) in the mineral contents of the pigmented rice.

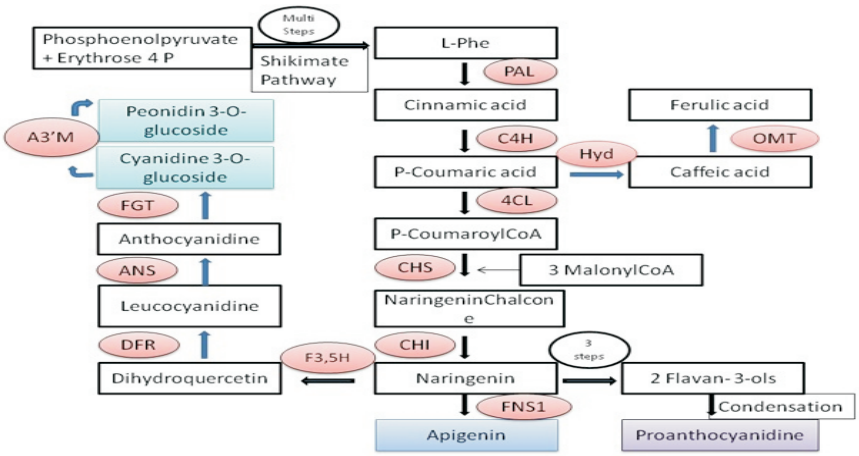

FIGURE 2. Schematic simplified diagram of anthocyanin and proanthocyanidine synthesis in pigmented rice. Enzymes :PAL- Phenylalanine ammonia lyase; C4H-Cinnamate- 4- hydroxylase; 4CL- Coumarate CoA Ligase; CHS-Chalcone synthase; CHI-Chalconeisomerase. FNS1:Flavone synthase-1; F3,5H: Flavone 3'5'Hydroxylase; DFR: Dihydroflavonol 4 reductase; ANS:Anthocyanidine synthase; FGT:UDP-glucose flavonoid 3-O-glucosyl transferase; A3'M: S adenosyl methionine transferase.

Constraints of popularization for pigmented rice and future scope

Grain quality and yield traits are most likely quantitative in nature genetically and expected to be influenced by genetic constitution of the plants, environment fluctuation and the genotype × environment interaction. The grain yield of some Indian pigmented rice was 15 to 16 g / plant which is lower than white rice. The head rice recovery of many pigmented rice are lower than standard value (>60%). Due to lower HRR, the farmers are not generally adopted those varieties. Many pigmented rice varieties have lower amylose content and generally categorised as waxy rice. Therefore, boiled rice of these varieties/cultivars is very soft in nature and the consumers do not prefer it in many states of India though it is highly popular in north eastern states of India.In India, pigmented rice are widely utilised for preparation of sweet dishes and other ethnic foods. It was observed that the pigmentation of rice is mainly associated with bran layers. Therefore, after milling or polishing of the grains, maximum amount of pigmentation is removed; only very little or no pigmented bran is adhered with the endosperm, which is generally white in colour. However pigmented endosperm rice varieties are rarely available in the world. Some black and red rice cultivars are also unsuitable for cultivation due to higher plant height and longer duration as compared to most of the popular rice varieties. However, improvement of grain and physiological quality of pigmented rice can be done through conventional breeding or modern biotechnological tools like gene editing or recombinant technology.

CONCLUSION

Pigmented rice is nutritionally rich but in many aspects they are unsuitable for farmers and consumers point of view. Many pigmented rice are the good source of phenolics, flavonoids, minerals, fibers, olis and proteins but after milling a substantial amount of these nutrients are removed with the by-products like bran. On the other hand, due to lower amylose content, breaking of the grain is occurred at the time of milling. So, it is the challenge to the scientist to upgrade the pigmented rice cultivars with minimising the unwanted traits, related to physiology and grain quality. However, as pigmented rice is superior in term of nutritional quality and having export potential, the whole rice i.e. brown rice can be utilized for consumption as well as for commercial purpose. Therefore, research should be initiated for preservation of the whole rice, which is susceptible to rancidity.

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