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Bioactive phytochemicals in Rice Bran: processing and functional properties

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Abstract : Rice bran is a by-product of rice milling industry and constitutes around 10% of the total weight of rough rice. It is primarily composed of aleurone, pericarp, subaleurone layer and germ. Rice bran is a rich source of vitamins, minerals, essential fatty acids, dietary fiber and other sterols. The quantification of γ -oryzanol in Rice bran can be performed by many methods that involve extraction of Rice bran oil from the bran, followed by analysis of the amount of γ oryzanol in the Rice bran oil by HPLC. In order to determine the amount of γ -oryzanol in Rice bran oil it is very important to completely extract this fraction from the oil. Various extraction techniques have been used for the analysis of γ -oryzanol in Rice bran oil such as liquid–liquid extraction, solid phase extraction, supercritical fluid extraction (SFE) and direct solvent extraction. There is a widespread scientific agreement on various health benefits associated with consumption of dietary fiber. Consumer attitude towards health foods is promising and the scope of functional foods is growing in the world markets; rice bran is finding increased applications in food, nutraceutical and pharmaceutical industries. In addition to the physiological benefits provided by high fiber foods, studies have shown that fiber components can give texture, gelling, thickening, emulsifying and stabilizing properties to certain foods. Considering the importance of rice bran, this review aims to focus on the functionalities of rice bran, its health benefits and potential applications in food industry.

Key words: Rice bran, stabilization, Functional food, Health benefits.

1. Introduction

Rice bran is the by-product obtained during milling of rice grain which contains 12 to 15% protein^{1.} Rice bran protein is a good source of well-balanced amino acid, bioactive phytochemicals such as γ -oryzanol, tocopherols, tocotrienols contains² and hypoallergenic protein which is desirable in infant food formulation³. Rice bran is an abundant by-product from rice production which can be served as a low cost attractive feedstock for the production of bioethanol^{4,5}. Pure rice bran that was dilute acid pretreated enzyme saccharification has been reported as an effective substrate for ethanol production by Zymomonas mobilis biofilm. Z. mobilis biofilm has illustrated its potential for ethanol production from rice bran hydrolysate than free cells by representing higher survival, higher metabolic maintenance and higher ethanol yield when it is exposed to the toxic inhibitors⁶. Therefore, using biofilm as a biocatalyst represented its feasibility for ethanol production from lignocellulosic material which could lead to the reduction in the operating costs of bioethanol and minimizing the complexity of the process. Rice bran protein has enormous health benefits and good potentials in food industry applications⁷. Rice bran protein concentrate and isolate are not commercially produced due to lack of commercially feasible extraction method⁷. Fabian and Ju, (2011) reported that several treatment methods (physical, heat, enzymic and chemical) have been used in the extraction of protein from rice bran. The authors further reported that the use of enzyme and subcritical water treatment showed promising protein yield among all the available pretreatment methods, but the relative expensive cost of enzyme and the quality of protein extracted at relatively high temperatures for subcritical water treatment constitutes limitations to the use of the methods due to reduced protein functionality⁷.

Shih (2003) found that treatment of rice bran by chemical, enzyme and heat methods prior to protein extraction could affect, often adversely, the protein functionality⁸. Rice bran is mostly burnt off at the rice mills and very little is used in animal feed. By understanding functional properties of dietary fiber, one can increase its use in food applications and aid in developing food products with high consumer acceptance. By continuous research on properties of rice husk ash, the results shows that it contain high silica content which is more than 90%, it reduces shrinkage cracks and leads to increase the strength of concrete. The many researchers are done research on rice husk ash and they presented their results of modified concrete properties⁹.

We systematically studied the chemical constituents and functional properties of dietary fiber from rice bran¹⁰. Rice bran hemicellulose B (RBHB) had been reported to have many biological activities including decreasing blood cholesterol and preventing colon cancer¹¹. This review describes the functionalities and health benefits of rice bran. Supplementation of rice bran in various foods is also discussed.

2. Industrial processing of rice bran, in order to obtain phytochemical-rich fractions

 γ -Oryzanol is an important value-added co-product of rice bran processing. Therefore, research to improve the recovery of γ -oryzanol and other phytochemicals, in order to obtain fractions enriched in a particular compound or group of compounds, has been conducted. Within this concern, particular attention has been paid to brown rice milling and Rice bran oil extraction and refining by either physical or chemical techniques. Relatively novel procedures involve the use of SC–CO₂, subcritical water and enzymes¹². Studies to characterize beneficial properties on these metabolites are highly recommended in order to convert this magnitude of biodiversity into the continuous innovation profitable for human living^{13,14}.

2.1. Influence of milling

Rice bran has been traditionally processed as a homogenous material; however, it has been shown that high-value components in the rice bran layer vary according to kernel thickness, bran fraction, rice variety, and environmental conditions during the growing season¹⁵. Thus, current rice milling technology produces rice bran from different layers of the kernel caryopsis. The phytochemical contents of each of these fractions can vary widely, the γ -oryzanol concentration being higher in the outer bran layers^{16,17,18}. Rice bran fractionation is advantageous for two reasons: (1) some fractions contain higher concentrations of components of interest, with respect to the overall bran layer average, and (2) less bran needs to be processed to obtain components of interest¹⁵.

According to Ha et al. (2006), the lipid, tocols, γ -oryzanol, squalene and octacosanol contents of both brown and milled rice decreased as the degree of milling increased; however, the phytosterol profile remained the same, β -sitosterol being the most abundant (50–56% of total phytosterols). Milled and brown rice showed also differences in the relative percentages of α -tocopherol, and α and γ -tocotrienols¹⁹.

2.2. Chemical and physical refining of rice bran oil

 γ -Oryzanol and most other phytochemicals are largely lipophilic, and thus are extracted with the rice bran oil; however, differently from tocols, γ -oryzanol is transferred to soapstock during the neutralization step of chemical refining of RBO²⁰. Alternatives to conventional chemical refining, including physical refining and the use of membrane technology²¹ have been proposed. Zigoneanu et al., (2008) have investigated the microwave-assisted extraction of rice bran with isopropanol and n-hexane at increasing temperatures. The increase in tocols with temperature, from 40 to 120 °C, was 59.6% for isopropanol and 342% for n-hexane; however, isopropanol was better than nhexane for the extraction of γ -tocopherol and γ -tocotrienol, also leading to higher oil yields at high temperatures. Further, fractions extracted with isopropanol at 120 °C had the highest antioxidant activity²². Van Hoed et al. (2006) examined the effects of each individual step of chemical refining of RBO on its major and minor components. Large γ -oryzanol losses and a change in the individual phytosterol composition were produced by either alkalinisation or neutralisation. After bleaching, some isomers of 24-methylenecycloartanol were detected. Due to their relatively high volatility, free phytosterols and tocotrienols were stripped off from RBO during deodorisation, and thus concentrated in the deodoriser distillate, but the RBO γ -oryzanol concentration did not change upon deodorization²³.

2.3. Extraction with supercritical-CO₂

Owing to the low viscosity and high diffusivity of SC fluids, highly efficient extraction procedures can be developed. Further, from the environmental viewpoint, SC–CO₂ is much better than organic solvents. Xu and Godber (2000), compared liquid organic solvents with SC–CO₂ relative to efficiency for extracting lipids and γ oryzanol from rice bran. Among the solvents tested, a 50:50 n-hexane/isopropanol mixture at 60°C for 45–60 min produced the highest γ -oryzanol yield. Without previous saponification, the yield of γ -oryzanol was approximately two times higher than that with saponification. However, using SC–CO₂ the yield of γ -oryzanol was approximately four times higher than the highest yield obtained by extraction with liquid organic solvents²⁴.

2.4. Extraction with subcritical water

At temperatures and pressures close to that of its critical point (374 °C, 22 MPa), water behaves like a highly hydrophobic solvent, thus being useful to extract lipophilic substances from solid and semi-solid matrices. After cooling and depressurising, a lipophilic and a hydrophobic fraction can be obtained. High recoveries in extraction times much shorter than using SC–CO₂ can be achieved. Today, however, subcritical water extraction for rice bran processing has been still scarcely investigated, and as far as we know, scaled-up processes of industrial interest have not yet been described. Hata et al., (2008) have also treated defatted rice bran with subcritical water in the 180– 280 °C range. The total sugar concentration was the highest for the extracts at 200 °C. The protein concentration and radical-scavenging activity increased at increasing temperatures. The extracts obtained below 200 °C showed emulsifying and emulsion-stabilising activities^{25, 26}.

2.5. Enzymatic processing techniques

The use of enzymes in rice bran processing, including enzymes specifically modified by genetic engineering, is still today a new and relatively unexplored technology. Potential applications are the development of improved food products and novel products of pharmaceutical interest. A processing technology to polish rice in a selective way with the help of xylanases and cellulases has been developed^{27,28}.

Enzymes produced by *Aspergillus* sp. And *Trichoderma* sp. acted upon the non-starch polysaccharides of the bran layers of moistened brown rice, releasing their monomeric sugar constituents, as detected through HPLC. Surface degradation of the rice grain was also studied by scanning electron microscopy²⁸. Selective degradation of bran layers facilitated the retention of phytochemicals. Antioxidant activity followed the order brown rice > enzyme-treated rice > milled rice. In comparison to mechanically-milled rice, bio-polished rice had better cooking attributes and higher antioxidant concentrations²⁷.

3. Stabilization of rice bran

Although being an excellent nutrient source, rice bran is not suitable for human consumption due to the rancidity caused by presence of lipases. While removing bran layers from the en- dosperm during milling, the individual cells are disrupted and lipase enzymes come into contact with fat causing hydrolysis to free fatty acids (FFA) and glycerol²⁹. Additionally, various antinutritional factors present in rice bran limit its use as a food ingredient. The factors include trypsin inhibitors, heamagglutinin–lectin and phytates^{30,31}. However, stabilization, an enzyme inactivation process, is widely employed to extend the shelf life of rice bran, enabling incorporation of rice bran back into our diet. Studies have shown that all undesirable factors except phytates present in bran are proteinicious in nature, therefore, mild acid and alkali treatment and thermal cooking can denature these proteins³¹. Different techniques are employed for rice bran stabilization (Table1).

| Stabilization technique | Conditions | |
|---------------------------------|---|--|
| Hot air drying | 100 °C for 1h | |
| Steaming | 100 °C for 30 min | |
| Refrigeration | 2 °C | |
| Sun drying | 47 °C (maximum) 7 h per day for 2 days | |
| Fluidized bed drying | 84 °C for 1 h | |
| Chemical stabilization Spraying | 1000 ppm HCl solution | |
| Microwave heating | 2450 MHz for 2 min | |
| Infrared | 700 W for 30 min | |
| Ohmic heating | 20–40% moisture; 44–72 V/cm voltage gradients | |

Table 1. Various techniques for stabilization of rice bran³².

4. Health benefits of rice bran

Natural products obtained from plants have been used as a prominent source of prophylactic agents for the prevention and treatment of diseases in humans and animals³³. Nutraceuticals including phytochemicals are perceived as offering some of the greatest opportunities for improving human health³⁴. Phytochemicals of dietary and non-dietary origin have been the focus of researchers in the recent past because of their potential to counter various diseases. Rice bran contains phytochemicals with promising health benefits³⁵. Some of the important bioactive components presented in rice bran are presented in Table 2. Rice bran oil rich in natural antioxidants may play a role in reducing the risk of chronic diseases³⁵.

| Anthocyanins and flavonoids | Polymeric carbohydrates | Phenolic and cinnamic acids | Steroidal compounds |
|--|----------------------------|-----------------------------|--|
| Anthocyanin monomers, dimmers and polymers | Arabinoxylans | Caffeic acid | Acetylated steyrl glucosides |
| Apigenin | Glucans | Coumaric acid | Cycloartenol ferulate |
| Cyaniding glucoside | Hemicellulose | Catechins | Campesterol ferulate |
| Cyanidin rutinoside | | Ferulic acid | 24- methylenecycloartenol ferulate |
| Epicatechins | | Gallic acid | γ-oryzanol |
| Eriodtyol | | Hydroxybenzoic acid | β-sitosterolferulate |
| Hermnetins | | Methoxycinnamic acid | Tocopherols |
| Hesperetin | | Sinapic acid | tocotrienol |
| Isohamnetins | | Syringic acid | |
| Luteolin | | Vanillic acid | |
| Peonidin glucoside | | | |
| Tricin | | | |

Table 2. Bioactive compounds present in rice bran³⁷.

The possible mechanisms for the hypocholesterolaemic effect of rice bran oil and its effect on aortic fatty streak (early atherosclerosis) formation was investigated. For this experiment, 30 hamsters were fed, for 8 weeks, chow-based diets plus 0.03-0.05% added cholesterol and 5-10% (w/w) physically-refined rice bran oil. Both plasma total cholesterol and LDL cholesterol were significantly reduced by the supplementation. A significant 30% increase in neutral sterol excretion was reported. The results suggested that lipid lowering is associated with attenuated cholesterol absorption and subdued fatty streak formation with this oil may be associated with its non-triacylglycerol components³². It was concluded that oryzanol and rice bran oil diets have significantly lower aortic cholesteryl ester accumulation compared to the control diet³⁹.

Cholesteryl ester is a highly hydrophobic ester with a role in atherosclerosis. Liquid-phase of this ester constitutes dominant component of atherosclerotic plaque and leads to atherothrombosis^{40,41,42}. The effect of γ -

oryzanol on lipid metabolism in Wistar rats with type 2 diabetes was evaluated. 15% palm oil added with 5.25g γ -oryzanol markedly reversed the increase in plasma LDL cholesterol, plasma triacylglycerol and hepatic triacylglycerol levels⁴³.

Fractions of peptides were isolated and characterized from rice bran for possible inhibitory effects against obesity. Hydrolysis and gastrointestinal juices treatment followed by fractionation resulted in a pentapeptide with anti-obesity effects⁴⁴. The antioxidant capabilities of anthocyanin and tocol extracts from black rice bran were evaluated. In an emulsion system containing cholesterol, anthocyanin proved superior in stabilizing cholesterol, while tocol showed better inhibition of fatty acid oxidation⁴⁵.

The effect of vitamin E analogues, especially γ -tocotrienol on hepatic triglyceride deposition in rat hepatocytes, was investigated and its inhibitory role was established. It was suggested that the tocotrienol might prevent hepatic steatosis and ameliorate endoplasmic reticulum stress and subsequent inflammation in the liver⁴⁶. Whether rice bran enzymatic extract-supplemented diet is capable of attenuating microvascular alterations in obese rats was assessed. The extract (1% and 5%) administration for 20 weeks restored microvascular function in obese rats through a marked increase in nitric oxide and endothelial-derived hyperpolarizing factor contribution by up-regulation of eNOS and calcium-activated potassium channel expression, respectively. Also, the modulation caused substantial reduction of microvascular inflammation and superoxide anion formation⁴⁷.

5. Supplementation of rice bran for development of functional foods

A number of studies have been carried out to evaluate rice bran as a functional ingredient in various foods to improve the nutritional quality. Rice bran being high in dietary fiber and in view of its therapeutic potential, its addition can contribute to the development of value-added foods or functional foods that currently are in high demand⁴⁸. Rice bran hemicelluloses and preparations from defatted rice bran have great potential in food industry, especially in development of functional foods such as functional bakery products¹⁰. Enrichment of bakery products with rice bran and its effects are summarized in Table 3.

| Product enriched | Purpose of addition | Inference |
|--|--|---|
| Gluten-free bread with dietary fiber fractions | Quality improvement | Acceptable structural and textural quality Sensory acceptance increased and shelf life extended⁴⁹ |
| Cookies with rice bran | Effect on cooking quality | Average width, thickness and spread factor increased with increment in rice bran⁵⁰. Supplementation of heat stabilized rice bran at 10% is suitable for production of rice bran supplemented cookies⁵¹ |
| Pasta | Effect of enrichment on the color, cooking, sensory quality and shelf life of enriched pasta | Pasta with added brans had higher dietary fiber and protein contents⁴³ Rice bran up to 15% level did not affect the physico- chemical, cooking and sensory quality of pasta. |

Table 3. Enrichment of bakery products with rice bran⁴⁸.

6. Conclusion

Rice bran is a good source of proteins, minerals, fatty acids, fiber and fatty acids. Considering the importance of rice bran, it can serve as an important raw material for the development of nutraceuticals and functional foods including bread, corn flakes, ice cream, pasta, noodles and zero-trans-fat shortening. Owing to numerous health benefits associated with the consumption of rice bran, detailed in vivo studies are recommended to create a strong data base. Also, the comparative analysis of the shelf life achieved by stabilization with different techniques has not been carried as of now which is an interesting area of research.

This can help predict the best procedure for stabilization to enhance the supplementation of rice bran in various food systems.

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