Review

Rice Bran Oil: Emerging Trends in Extraction, Health Benefit, and Its Industrial Application

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Abstract: Rice bran oil (RBO) is unique among edible vegetable oils because of its unique fatty acid composition, phenolic compound (γ-oryzanol, ferulic acid) and vitamin E (tocopherol and tocotrienol). It has become a great choice of cooking oil because of its very high burning point, neutral taste and delicate flavour. Non-conventional methods of RBO extraction are more efficient and environmentally friendly than conventional extraction methods. Advances in RBO extraction using innovative extraction strategies like super/sub-critical CO2, microwave-assisted, subcritical H2O, enzyme-assisted aqueous and ultrasound-assisted aqueous extraction methods have proven to significantly improve the yields along with improved nutritional profile of RBO. The compositions and strategies for stabilization of RBO are well discussed. The constituents are present in the RBO contribute to antioxidative, anti-inflammatory, antimicrobial, anti-diabetic and anti-cancerous properties to RBO. This has helped RBO to become an important substrate for the application in food (cooking oil, milk product and meat product) and non-food industries (polymer, lubricant, biofuel, structural lipid and cosmetic). This review provided comprehensive information on RBO extraction methods, oil stabilization, existing applications and health benefits.

Key words: rice bran oil; extraction; functionalization; stabilization; food/non-food application; fatty acid

Rice (Oryza sativa L.) is widely consumed as cereal crop worldwide, majorly in Asia and Africa, and among all countries, China and India are the largest exporters, which account for approximately 50% of rice trades (FAO, 2019). Rice is a good source of carbohydrates, minerals (calcium and iron) and vitamins (thiamine, pantothenic acid, folate and vitamin E), when compared with maize, wheat and potato. Along with white rice, some specialty coloured and aromatic rice varieties are also cultivated. Anthocyanins in the bran layer are responsible for the rice colour, and these coloured rice varieties are rich in antioxidants and functional components (Priya et al, 2019). Milling of rice yields, 70% of rice (endosperm) as the major product and by-products, consists of 20% husk, 8% bran and 2% germ (van Hoed et al, 2006). In recent years, rice bran has been explored for its potential biological functionalities, which include antioxidant properties and anti-inflammatory activity, reduced incidence of cancer, prevent coronary heart diseases, and decreased cholesterol level. One of other important component present in rice bran is phytic acid (59.4 to 60.9 g/kg) (Liu et al, 2005; Canan et al, 2011; Kumar et al, 2020). Scientific documentation on phytic acid indicates its beneficial effects on human health, especially in preventing the renal calculi, cancer, diabetes, Parkinson’s disease, and hypolipidemic effect (Al-Fatlawi et al, 2014; Barahuie et al, 2017; Masunaga et al, 2019).

Rice bran is obtained from outer layer of the brown (husked) rice kernel during milling. It has emerged as
potential by-product from rice processing industry due to the increasing demand for rice bran oil (RBO) as healthy functional food ingredients. Rice bran contains 18%–22% oil, including an array of bio-active phytochemicals such as oryzanol, phytosterol, tocotrienol, squalene, polyicosanol, phytic acid, ferulic acid and inositol hexaphosphate (Khatoon and Gopalakrishna, 2004; Ardiansyah et al, 2006; Saikia and Deka, 2011). Ferulic acid is the major phenolic acid, followed by p-hydroxycinnamic acid, sinapinic, gallic, protocatechuic, p-hydroxybenzoic and vanillic acid in rice bran (Goufo and Trinade, 2014). RBO contains a range of fatty acids, with 47% monounsaturated, 33% polyunsaturated and 20% saturated. These phytochemicals show numerous biological activities by acting as antioxidant, anti-diabetic, anti-carcinogenic, antiatherogenic and antihyperlipidaemic agents (Garofalo et al, 2021). Bio-composites based on low density polyethylene (LDPE) and acrylic acid grafted polyethylene are developed using rice bran as solid carrier and tea tree oil for achieving high antimicrobial efficiency with sustained release of bioactives from the blends. LDPE-based polymer composite with 30% of rice bran improves the mechanical properties of the packaging film in terms of hardness value (El-Wakil et al, 2020).

Advances in RBO extraction strategies, like super/sub-critical CO₂, microwave-assisted subcritical H₂O, and ultrasound-assisted aqueous extraction methods, help to make RBO as an important substrate for the food and non-food industries. Non-conventional methods of RBO extraction are more efficient and environmentally friendly than conventional extraction methods. This review comprehensively discussed various non-conventional techniques for the extraction of RBO, followed by its functionality enhancement by stabilization strategies, and also presented various industrial application of RBO along with the health benefits in human body.

RBO extraction methods

RBO is a rich source of free fatty acids (FFA), waxes, unsaponifiable constituents (4.3%) and polar lipids, and the extraction process is challenging. Extraction methods including cold pressing (Mingyai et al, 2017; Wongwaiwech et al, 2019), Soxhlet extraction (Mingyai et al, 2017; Wongwaiwech et al, 2019), supercritical CO₂ extraction (SC-CO₂) (Mingyai et al, 2017) and microwave extraction (Zigoneanu et al, 2008) have been studied (Table 1).

Conventional method

Conventional methods mainly use solvents and cold pressing techniques for extraction of RBO (Fig. 1).

Solvent extraction method

Solvents are employed to recover oil from seeds with low oil content, or from pre-pressed oil cakes in order to obtain high oil content (Fig. 1-A). Commercial-grade hexane is the solvent of choice throughout the world for economic reasons. It is excellent oil solvent in terms of solubility and ease of recovery. However, hexane is culprit of many complications. It produces poor colour quality RBO and also responsible for serious environmental problems (air pollution), in addition to other health hazards due to its toxicity (Lucas, 2000). Over the years, many attempts have been made to find alternative solvents, in particular alcohols, halogenated hydrocarbons, hydrocarbons, carbon dioxide (supercritical fluid extraction) and even water (Garofalo et al, 2021). Some short chain alcohols such as ethanol and isopropanol have also been proposed as an alternative solvent for extraction because of their greater safety (Patel and Naik, 2004; Zhang et al, 2008). d-limonene, an agricultural by-product from the citrus industry, is also a viable alternative to hexane-based solvents to achieve high yields of crude edible oil with good quality (Liu and Mamidipally, 2005). However, potential use of limonene as a solvent for edible oil extraction is the problem of high-energy cost that is mainly due to high-boiling point and slightly high latent heat of limonene. Energy-saving solvent recovery technology, such as membrane separations, might eventually overcome this obstacle and render limonene-based oil extraction affordable. An attempt was made to replace hexane with ethanol and isopropanol to extract RBO (Capellini et al, 2017) under different solvent hydration degrees (0%, 6% or 12%), using rice bran to solvent ratio (3:1) at different temperatures (50 °C to 80 °C) for 1 h. The maximum RBO extraction yield (160 g/kg) was obtained using ethanol and isopropanol at 80 °C, while the highest γ-oryzanol yield was obtained using ethanol with 6% of water, regardless of the temperature.

In another study, same yield of RBO (170.9 g/kg) was obtained by using isopropanol and hexane, which is higher than the yield by using ethanol after 4 h of extraction using a Soxhlet apparatus. Furthermore, higher γ-oryzanol concentration (2.609 mg/kg) was observed when RBO extracted with ethanol than hexane (2.229 mg/kg) or isopropanol (2.094 mg/kg) (Shukla and Pratap, 2017).

Cold pressing method

Mechanical pressing (cold pressing) method does not
Cold pressing can produce RBO with superior quality (Pre, 2008). Mild heating followed by filtration to get clean RBO. Pre-treatment of rice bran with low temperature before cold pressing can produce RBO with superior quality leaving 8%–14% of the available oil in the cake.

Cold pressing uses mechanical screw press with mild heating followed by filtration to get clean RBO. Pre-treatment of rice bran with low temperature before cold pressing can produce RBO with superior quality (Srikaeo and Pradit, 2011). A combination of short cooking time and cold pressing is more effective and efficient. A combination of ultra-sonication heating pre-treatment followed by mechanical pressing enhances the quality and extraction rate of RBO (Phan et al, 2019). This combination brings changes in structure, fissures or cavities in the rice bran fractions, improving the mass transfer and oil extraction efficiency (Kurian et al, 2015).

Table 1. Optimized conditions and parameters of RBO extracted by various conventional and non-conventional methods.

| Method                        | Input factor/parameter | Optimized condition | Finding                                                                 | Reference               |
|-------------------------------|------------------------|---------------------|-------------------------------------------------------------------------|-------------------------|
| Soxhlet extraction method     | Temperature            | 65 °C               | Solvent extraction using hexane as the solvent is an effective method for RBO extraction | Amarsinghe and Gangodavilaghe, 2004 |
|                              | Solvent                | Hexane              |                                                                         |                         |
| MAE method                    | Time                   | 30, 60, 90 and 120 s per step | MAE is a promising approach for extracting RBO with superior quality oil, an increase in oil yield and reduction in extraction time | Pandey and Shrivastava, 2018 |
|                              | Power                  | 300, 500, 700 and 900 W |                                                                         |                         |
|                              | Solvent used           | Hexane              |                                                                         |                         |
|                              | Solid to liquid ratio  | 1:1                 |                                                                         |                         |
|                              | Time                   | 60 min              | Methanol is the most effective solvent for extraction of γ-oryzanol containing RBO | Kumar et al, 2016       |
|                              | Power                  | 800 W               |                                                                         |                         |
|                              | Solvent used           | Petroleum ether, hexane and methanol |                                                                         |                         |
|                              | Solid to liquid ratio  | 1:3                 |                                                                         |                         |
| UAE method                    | Time                   | 60 min              | Ultrasound treatment significantly boosts the γ-oryzanol extraction capability of methanol as compared to conventional extraction method | Kumar et al, 2016       |
|                              | Power                  | 20 kHz              |                                                                         |                         |
|                              | Solvent used           | Petroleum ether, hexane and methanol |                                                                         |                         |
|                              | Frequency              | 24 kHz              |                                                                         |                         |
|                              | Temperature            | 38 °C               |                                                                         |                         |
| Supercritical CO₂ extraction method | Time               | 30 min              | Supercritical CO₂ may be focused on minimizing operational costs and maximizing oil yields | Juchen et al, 2019      |
|                              | Pressure               | 100, 150 or 200 bar |                                                                         |                         |
|                              | Temperature            | 40 °C, 60 °C or 80 °C |                                                                         |                         |
|                              | Solvent used           | Ethanol             |                                                                         |                         |
|                              | Solid to liquid ratio  | 0:1, 0.5:1, 1:1, 2:1 |                                                                         |                         |
| Subcritical water extraction method | Time                | 10–20 min           | Subcritical water extraction is an environmentally friendly technique for lipase inactivation and stabilization of RBO | Pourali et al, 2009   |
|                              | Temperature            | 180 °C–240 °C       |                                                                         |                         |
|                              | Solvent used           | Deionized water     |                                                                         |                         |
|                              | Solid to liquid ratio  | 1:6                 |                                                                         |                         |
| Subcritical CO₂ extraction method | Temperature         | 27 °C–29 °C         | Subcritical CO₂ extraction method increases approximately 10 times more oryzanol and tocol compounds and has lower free fatty acid levels and peroxide values compared with conventional method | Chia et al, 2015       |
|                              | Pressure               | 68–70 bar           |                                                                         |                         |
|                              | Solvent used           | Deionized water     |                                                                         |                         |
|                              | Solid to liquid ratio  | 1:24                |                                                                         |                         |

MAE, Microwave-assisted extraction; UAE, Ultrasound-assisted extraction; RBO, Rice bran oil; SC-CO₂, Supercritical CO₂.
techniques to improved non-conventional techniques such as supercritical fluid extraction (SFE), ultrasound-assisted extraction (UAE), microwave-assisted extraction (MAE) and enzyme-assisted aqueous extraction (EAAE) (Fig. 2).

**Supercritical CO\(_2\) extraction (SC-CO\(_2\))**

SFE provides a range of benefits including no risk of solvent contamination, as well as offering routes to overcome some of the limitations in conventional extraction. Compared with organic solvents extraction, SFE is faster and more efficient because of higher penetration power into the matrix and beneficial transport properties (Table 1).

An interest in SC-CO\(_2\) extraction has grown over the past few years (Fig. 2-A). It refers to separation technology which uses supercritical fluid i.e. CO\(_2\) above the critical temperature and pressure, making it an attractive solvent for temperature-sensitive materials (Sookwong and Mahatheeranont, 2017). To increase the effectiveness of extraction, CO\(_2\) is added with polar substances to improve the solubility and extraction rate (Santos et al, 2017). SC-CO\(_2\) along with ultrasound may be used to improve the dynamics of the RBO extraction. SC-CO\(_2\) extraction method yields 7.00%–9.60% of RBO. Oryzanol precursors (campesterol, β-sitosterol, stigmasterol and 4-methylenecycloartenol) were identified in RBO using the SC-CO\(_2\) + ultrasound extract method and claimed that this combination is a promising approach for the extraction of targeted functional compounds. Soares et al (2018) investigated the effects of SC-CO\(_2\) and ethanol (co-solvent) ratios. Juchen et al (2019) indicated that temperature of 40 °C and pressure of 200 bar result in overall high oil yields (25.48%) without residual solvent in RBO.

**Subcritical CO\(_2\) fluid extraction**

To overcome the disadvantages of SC-CO\(_2\) at high pressure to maintain solubility, a new approach recently emerged. The subcritical CO\(_2\) Soxhlet is an innovative technique which combines the advantages of Soxhlet and CO\(_2\) extraction to yield RBO (Chia et al, 2015). Extraction of RBO with supercritical and subcritical works under the same principle, subcritical CO\(_2\) operates with a temperature below 31.1 °C and CO\(_2\) pressure of 72.9 bar (Xuan et al, 2018). During this process, CO\(_2\) is continuously restored through boiling and later precipitation. Chia et al (2015) compared subcritical CO\(_2\) and hexane extraction method for RBO and reported that the yields using subcritical CO\(_2\) and hexane are 13.0%–14.5% and 22.0%, respectively. Using subcritical CO\(_2\) process, oil contains about 10 times more oryzanol and tocot compounds and has lower FFA levels and peroxide values compared with hexane-extracted oil.

**Subcritical water extraction (SWE)**

SWE offers a suitable, cost-effective and environmentally safe alternative compared to other methods as it takes advantage of the special properties of supercritical water under high temperature and pressure conditions (100 °C–374 °C, > 5 MPa) (Gbashi et al, 2017). The principle of SWE is based on the molecular structure and thermodynamic properties of water. The water under the pressure of 22.1 MPa and temperature below 374 °C is known as subcritical water (SW) (Zhang et al, 2020). When SW is used for extraction of functional compounds, the technique is known as SWE. The increased temperature results in an increase in its vapour pressure, mass transfer and diffusion, and decreases its surface tension and viscosity (Xu et al, 2016). SWE alters the dielectric constant (\(\varepsilon\)) of water by making favourable changes in the pressure and temperature (Gbashi et al, 2017). A simultaneous increase in temperature and pressure decreases \(\varepsilon\) of water to 25 from a normal of 80. This assists water to act like organic solvents and facilitates the extraction of desired bioactives with varying polarity. Temperature is the most important and determining factor in SWE, and it is used to control the extraction rate of analytes. And if other parameters of the extracting solvent temperatures as high temperatures, it is not suitable for extraction of heat sensitive compounds of oil, as well as subcritical water could be more reactive.
and corrosive compared to water at ambient conditions. The energy delivered by SWE has the potential to interfere with solute-solute and solute-matrix interactions, and results in decreasing the activation energy needed for desorption. To inactivate lipase and to stabilize RBO, SWE is an environmentally friendly method because of its green, simple and non-flammable nature. Pourali et al. (2009) reported a complete lipase enzyme decomposition and a maximum RBO yield of 249 mg/g (94% of the total oil from rice bran), which is comparable with the quantity obtained by means of hexane extraction. However, SWE has reported the wonderful results for the extraction of other oils, such as sunflower, soybean, cottonseed or jojoba oil (Ndlela et al, 2012; Ravber et al, 2015) and certain functional compounds, such as polyphenols, essential oils, carotenoids, flavonoids, and flavor and
fragrance compounds (Rozzi and Singh, 2002), but it is still not applied extensively for RBO extraction.

**UAE method**

Recently, UAE has been widely applied. Ultrasound is transmitted through a medium as a pressure wave and causes an excitation in the form of enhanced molecular motion (Fig. 2-B). Ultrasonication induces cavitation which increases the permeability of the plant tissues. Microfractures and disruption of cell walls provide more evidence for the mechanical effects of ultrasound, thus, facilitating the release of their contents (Liu et al, 2014). UAE has many advantages, including high extraction yield, high reproducibility, low solvent use, short extraction time, low running cost, limited environment impact and easy adaptation to industrial scale use (Vuong et al, 2014). UAE is mainly used for food processing purpose including drying, extraction and emulsification. Sonication (2 MHz) assisted extraction of RBO results the FFA and peroxide values below the industrial specification level as well as an enhancement in phenolic compounds (Martínez-Padilla et al, 2018). Khoei and Chekin (2016) employed response surface method to optimize the UAE parameters to obtain the maximum yield of RBO. The oil extracted by UAE had a lower content of FFA and lower colour imparting components than the hexane-extracted oil. Both extraction methods give higher percentage of oil from parboiled rice bran compared with raw rice bran.

**MAE method**

MAE serves as a green technique over conventional methods for extracting fats and oils (Fig. 2-C). This process has recently gained popularity due to its less extraction time, energy and solvent consumption and as a promising alternative to conventional solvent extraction method. The low specific heat of lipids makes them susceptible to this radiation, thus facilitating their solubility in the extractant (Pandey and Shrivastava, 2018). Kumar et al (2016) compared conventional extraction methods with UAE and MAE methods and found that methanol extracts around 96% of the oil in rice bran and (85.0 ± 0.2) mg/kg of γ-oryzanol. Crude oil and total phenolic contents of microwave treated methanolic extract (80%) are significantly higher than those of ultrasonication and conventional solvent extracts. Isopropanol coupled with MAE yields a higher RBO than hexane at 82 °C, 2.1 bar and 95 W with an extraction time of 30 min. Shukla and Pratap (2017) reported that the RBO extracted with isopropanol, under the same experimental conditions, contains 2.256 mg/kg of γ-oryzanol. For edible grade vegetable oil, crude oil is refined and processed further either chemically (to remove FFA with a large excess of sodium hydroxide) or physically (to remove FFA and deodorize with a water degumming step) to meet the standards of specifications. Moreover, MAE has low risks and no major safety issues as most extractions are generally carried out under atmospheric condition. In addition, there are some drawbacks and limitations associated with MAE. As discussed earlier, non-polar solvent should normally be discouraged as they are poor absorbents for microwave heating (Chan et al, 2011).

**EAAE method**

EAAE has emerged as an eco-friendly and emerging green technology, which facilitates the release of oil in aqueous extraction processes, while avoiding the use of organic solvents (Fig. 2-D). This process involves the treatment of oil-containing materials with cell wall-degrading enzymes in order to extract oil and other components under milder processing conditions (Hannoungjai et al, 2001). EAAE requires a lower energy and solvent consumption than conventional solvent extraction, and results in an excellent product, which does not require any further refining steps. The main enzyme classes exploited for RBO extraction are cellulase, pectinase and protease (Rosenthal et al, 1996; Sengupta and Bhattacharyya, 1996; Moreau et al, 2007). Amylase has been employed to facilitate the extraction of RBO. However, this approach yields only a 5% increase in oil recovery (Hernandez et al, 2000). Use of enzymatic treatment alone does not result in high yields. Oil yields reported to be high when rice bran was treated with cellulase and pectinase and then extracted with n-hexane (Sengupta and Bhattacharyya, 1996). Further to increase the yield, a combination of protease, α-amylase and cellulase obtains RBO recovery of 76%–78% of those normally obtained with conventional extraction methods (Sharma et al, 2001).

**RBO compositions**

Rice bran, an underutilized by-product of rice milling industry, represents 5%–10% of the total grain. It constitutes crude protein (11%–13%), oil (20%), dietary fibers (22.9%) including hemicelluloses, arabinogalactan, arabinoxylan, xylol glycan and raffinose, and also γ-oryzanol, vitamin-E and minerals (Table 2) (Pourali et al, 2009). RBO is a light, odourless pale yellow and translucent oil with a mild nutty flavour (Ali and Devaranjan, 2017). Indian Council of Medical Research, National Institute of Nutrition (India), World Health Organisation, American Heart Association, the Chinese
Cereals and Oils Association have considered RBO as a “healthy oil” due to its fatty acid profile and other constituents. RBO is one of the healthiest edible oils due to balanced source of saturated (SFA: 20% palmitic acid), monounsaturated (MUFA: 42% oleic acid) and polyunsaturated fatty acid (PUFA: 32% linoleic acid) with an average ratio of 0.6:1.1:1.0, respectively (Lai et al, 2019). The typical composition of crude RBO is 81%–84% triacylglycerols (TAG), 2%–3% diacylglycerols (DAG), 1%–2% monoacylglycerols (MAG), 2%–6% FFA, 3%–4% wax, 0.8% glycolipids, 1%–2% phospholipids (PL) and 4% unsaponifiable fraction (Ghosh, 2007). The unsaponified fraction is rich in tocophenols and tocotrienols, γ-oryzanol, phytosterols, polyphenols and squalene. FFA, MAG and DAG in RBO are associated with enzymatic hydrolysis. As comparison to other vegetable oils, crude RBO tends to contain higher levels of non-TAGs, most of which are to be removed during refining processes.

Due to presence of tocopherols, γ-oryzanol and tocotrienols, RBO has several advantages over other cooking oils including oxidative stability and many health benefits. γ-oryzanol is a class of non-saponifiable lipids of RBO. It is a mixture of ferulic acid esters of triterpene alcohols and phytosterols (Rogers et al, 1993). Krishna et al (2001) investigated the effect of processing steps on the availability of γ-oryzanol. The results showed that 1% and 6% of γ-oryzanol are removed by degumming and dewaxing of crude RBO whereas alkali treatment removed up to 94% γ-oryzanol from original crude RBO. They further concluded that physical and alkali refined RBOs retain original amount γ-oryzanol of 1.10%–1.74% and 0.19%–0.20%, respectively. Pestana-Bauer et al (2012) reported γ-oryzanol of 12.4 g/kg in crude RBO whereas only 0.49 g/kg in purified RBO. Cardioprotective γ-oryzanol is exclusively found at high concentration in RBO, and therefore it is regarded as the heart-friendly oil. Besides, RBO also has vitamin E (both tocopherols and tocotrienols) and plant sterols including β-sitosterol, campesterol, isofucosterol and stigmasterol, which are antioxidants in nature (Zubair et al, 2012).

The qualitative characterization of RBO is generally performed using the Fourier transform infrared (FTIR) spectroscopy whereas confirmatory or quantitative profiling of saponifiable fatty acids/volatiles are done using the gas chromatography (GC) or GC-mass spectrometry (GC-MS). Different oils were studied using FTIR spectroscopy and observed that RBO has the close analogy with that of virgin olive oil with respect to the spectral peaks (Rohman and Man, 2012). FTIR spectra of RBO, olive oil and other plant oils were observed in the wavelength range of 650–4 000 cm⁻¹. Plant oils are mainly constituted by 90%–95% of the tri-acylglycerols along with some quantity of mono- and di-acylglycerols, hence, representing a similar FTIR spectra in all the tested plant oils. There are minor changes in FTIR spectral peaks of RBO and virgin olive oil. A more detailed study showed a difference in FTIR spectra in terms of small band shifts and of small changes in the relative peak intensity, especially at frequency region of about 1 117–3 007 cm⁻¹. The sterol composition of saponified fatty acid ester of crude RBO was analyzed using GC and GC-MS (Gunawan et al, 2006). GC-MS analysis showed that crude RBO is rich source of sitosterol, stigmasterol, citrostadienol, campesterol, cycloartenol and 24-methylenceycloartanol and the major fatty acids in RBO are oleic acid and linoleic acid.

### Rice bran and rice bran oil stabilization

RBO is considered as perfect and high-class cooking oil because of its low level of linolenic acid and its potential to lower serum cholesterol level. After milling, oil is very prone to degrade into FFA and glycerol by lipase activity, making it unfit for consumption. It is advised to stabilize rice bran or RBO immediately upon production. High FFA results in high refining loss, therefore, it is beneficial to have FFA level of less than 5% in the crude RBO, and exceeding amount of FFA (> 10%) is unfit for human consumption (Orthoefer and Eastman, 2005). Many efforts have made to improve the stabilization of rice bran by exploiting the principles of ohmic heating, microwave and infrared treatments. Ohmic heating at

| Table 2. Phytonutrients and fatty acid composition of rice bran oil. |
|------------------------|------------------|-----------------|
| Compound               | Composition (%)  | Reference       |
| γ-oryzanol             | 0.9–2.9          | Lloyd et al, 2000; Patel and Naik, 2004 |
| Tocopherol             | 0.10–0.14        | Lloyd et al, 2000; Patel and Naik, 2004 |
| Saturated fatty acid   | 22.5             | Orsavova et al, 2015 |
| Palmitic               | 21.6             | Krishna et al, 2006 |
| Stearic                | 2.1–4.7          | Orsavova et al, 2015 |
| Arachidic acid         | 1.0              | Krishna et al, 2006 |
| Myristic               | 0.30–0.39        | Krishna et al, 2006 |
| Monounsaturated fatty acid | 44.0             | Orsavova et al, 2015 |
| Oleic acid             | 42.6             | Krishna et al, 2006 |
| Palmitoleic acid       | 0.19             | Orsavova et al, 2015 |
| Polyunsaturated fatty acid | 33.6             | Orsavova et al, 2015 |
| Linoleic acid          | 28.0             | Krishna et al, 2006 |
| Linolenic acid         | 0.8              | Krishna et al, 2006 |
| n-3 Polyunsaturated fatty acids | 0.5             | Orsavova et al, 2015 |
| n-6 Polyunsaturated fatty acids | 33.1             | Orsavova et al, 2015 |
100 V/cm of electrical field strength and an alternating electric current of 60 Hz are applied for stabilization and enhancing extraction yield of RBO (Lakkakula et al, 2004). A high power and long exposure time effectively inactivate lipase. Convection microwave of output power of 700 W and frequency of 2 450 MHz were used at different power densities (2, 4 and 6 W/g) and times (1, 3 and 5 min) for arresting lipase activity to improve the storage and oxidative stability of RBO (Patil et al, 2016). Rice bran with microwave treatment of 4 W/g for 5 min is effective in limiting lipase activity up to 90 d. Irakli et al (2018) stated that infrared heating of rice bran at 140 ºC for 15 min is the most effective method for stabilization. Lavanya et al (2019) stabilized rice bran fractions by microwave heating continuously at 850, 925 and 1 000 W and 3.0, 4.5 and 6.0 min. They concluded that microwave heating at 925 W for 3.0 min is the suitable condition for RBO extraction and stabilization. For stabilization of rice bran and to check the residual lipase activities as an indicator of the stabilization efficiency, 11 different treatments (6 heating treatments and 5 non-heating treatments) were compared (Yu et al, 2020). The results indicated that microwave, extrusion, steaming, -80 ºC, ultraviolet irradiation, infrared heating have potential to stabilize rice bran. Extrusion and microwave processes are suitable for large-scale with short-time industrial processing although with increase of oil peroxide values and colour. However, ultraviolet irradiation process does not interfere with nutrients and oil quality, and is convenient and energy-saving stabilization method.

**Health benefits of RBO**

**Reduce oxidative stress and hypertensions**

Oxidative stress and hypertension may occur when there is an imbalance of free radicals in body and capacity of body to react to the negative effect through neutralization by antioxidant compounds. Considerable evidence suggests that high levels of reactive oxygen species induce cellular senescence, chronic degenerative diseases and diabetic complications (Lee et al, 2014). Medium chain fatty acid (MCLA)-rich RBO has potential to reduce the hydroperoxide formation by limiting the production of free radicals. Sengupta et al (2014) evaluated the antioxidative effects of capric, caprylic and lauric acid rich RBO and concluded that fatty acids rich RBO reduced lipid peroxidation. γ-oryzanol-rich RBO provides a protective mechanism against oxidative stress and hypertension. Treatment of virgin RBO mitigates all the harmful effects in hypertensive rats (Jan-on et al, 2020). Lisinopril (medication for hypertension) along with the virgin RBO is the most effective in controlling the symptoms of hypertension. The antihypertensive and lipid-lowering effect of consuming RBO is closely related with the activation of epithelial nitric oxide and suppression of angiotensin-converting enzyme, NADPH oxidase, and nuclear factor-κB pathways (Jan-on et al, 2020). Hence, the presence of antioxidative fatty acid and phenolic profile along with the modulating effect of RBO component ACE and other factors allows its potential use as anti-hypertensive and anti-oxidative agents.

**Anti-cancer**

Globally, cancer is a leading and common cause of mortality and morbidity, and many predictive risk factors are associated with anticancer medications. Inflammation and infection are an important inducer of tumour progression, and aid in cancer cell proliferation, and promote angiogenesis and cell mobility. Thus, reducing inflammation is a promising and potential target for treating cancer (Trinchieri, 2011). Forster et al (2013) reported functional ingredients and phytonutrients in rice bran are strongly associated with inhibiting human colorectal cancer cells. Recent data demonstrated that rice bran extracts and fermented rice bran play an important role in attenuating inflammation (Yu et al, 2019). γ-tocotrienol of RBO has potential to reduce pancreatic tumour growth by inhibiting the nuclear factor-κB (NF-κB)-mediated inflammatory micro-environment. Rice bran components act as anticancer agents by inhibiting the cell cycle between G0/G1 or G2/M and by promoting the apoptosis of the cancerous cells (Henderson et al, 2012). Researchers have established the positive roles of the various components of rice bran in prevention or slowing down the spread of gastric cancer, colon cancer, breast cancer, prostate cancer and blood cancer.

**Anti-diabetic activity**

Diabetes is a metabolic disorder in which glucose level in the blood increases due to inefficient or low levels of insulin. When insulin hormone is not produced in required amount, the level of sugar increases in the blood. An anti-diabetic potential and reduction in blood sugar was observed by Tantipaiboonwong et al (2017), who fed black and red rice extract for eight weeks to mice. Rice bran extracts show inhibitory activity against α-glucosidase, α-glucosidase activity and α-amylase,
and stimulate the glucose uptake by adipocytes (Boue et al., 2016; Saji et al., 2019). Bran extracts show insulin-like effects and positively correlated with the diabetes management. It manages diabetes by increased expression of GLUT1, reducing blood glucose and inhibiting the conversion of starch into glucose in the gut (Boue et al., 2016).

**Anti-inflammatory or anti-allergic agent**

Inflammation may lead to angiogenesis, promoting cell proliferation and cell mobility (Mantovani et al., 2008). Many epidemiological reports revealed the anti-inflammatory characteristics contributed by bioactive compounds (γ-oryzanol, cycloartenyl ferulate and ferulic acid) in RBO (Islam et al., 2008; Rigo et al., 2014). Dietary RBO provides cellular energy production by β-oxidation and modulates inflammatory behaviour in murine macrophages by modulating mitochondrial energy metabolism (Lee et al., 2019). RBO reduces the levels of pro-inflammatory cytokines i.e. interleukin-6 and tumour necrosis factor α, whereas anti-inflammatory interleukin-10 cytokine is upregulated in an in-vitro study on lipopolysaccharide stimulated RAW 264.7 murine cell lines (Lee et al., 2019).

Ferulic acid (4-hydroxy-3-methoxycinnamic acid), a component of RBO, possesses anti-inflammatory activity. Oka et al (2010) investigated the effect of cycloartenol ferulic acid ester (CAF), a natural product from RBO γ-oryzanol, in passive cutaneous anaphylaxis (PCA) reaction and mast cell degranulation. They concluded that CAF functions as an anti-allergic agent and prevents the binding of IgE antibody (play central role in allergic reactions) and FceRI, and attenuates mast cell degranulation, which is a main event in type I allergy. Islam et al (2014) reported that NF-κB-related inflammation has a major role in the late phase responses of allergy. γ-oryzanol shows an anti-allergic effect to inhibit the allergy by reducing the action of NF-κB.

**Anti-hypercholesterolemia and prevents cardiovascular diseases (CVDs)**

CVD, a most common disease of heart and blood vessels, is documented as the leading health threats worldwide (Goyal et al., 2018). Hyperlipidaemia or high cholesterol is a highly predictive risk factor for high blood pressure, blood sugar, CVD, liver damage and atherosclerosis. Numerous studies have reported phytosterols, oryzanols and tocotrienols in RBO have specific hypocholesterolaemia effects. γ-oryzanol is associated with reduction in plasma and serum cholesterol and decreased cholesterol absorption, resulting in treating hyperlipidaemia (Zavoshy et al., 2012; Saji et al., 2019). Chen and Cheng (2006) reported that phytosterols in RBO may reduce the absorption of cholesterol, thus inhibiting the cholesterol movement into micelles and reduced uptake in the intestines. Bumrungpert et al (2019) explained that consumption RBO rich in γ-oryzanol at the concentrations of 4 000, 8 000 and 11 000 mg/kg may ameliorate CVD risk factors by decreasing bad cholesterol levels and increasing antioxidant potential in hyperlipidaemic issues. The reduced cholesterol levels are attributed to the inhibition of key regulatory enzyme of the cholesterol biosynthesis pathway i.e. 3-hydroxy-3-methyl-glutaryl-CoA reductase popularly known as HMG-CoA reductase in liver by RBO. It was also speculated that γ-oryzanol reduces the absorption of the cholesterol or it may decrease the blood lipid by inhibiting the activity of cholesterol 7-alpha-hydroxylase (maintain cholesterol levels) (Bumrungpert et al, 2019). Above-mentioned reasons make RBO as a potential candidate to prevent CVDs in human beings.

**Immuno-stimulation effects**

Nutrition and nutritional status have a significant effect on immune functions and resistance to infection. According to this, many nutrients can manage the immune function, and lesser and higher consumption affects the activities of immune cells. Many clinical trials have demonstrated that an improvement in depressed immune functions is shown by using RBO (Park et al., 2017). Rice bran contains immune system boosting components including phytosterols, sterolins, γ-oryzanol, omega-3 fatty acids, phytonutrients and minerals (Park et al, 2017). The bioactive peptides prepared using enzymatic hydrolysis of rice bran demonstrated immune-modulatory effects along with antioxidant and anti-cancer activities. Rice bran dietary fibers, arabinoxylan and β-glucan have immune-stimulatory effect. MGN-3 (arabinoxylan) shows anti-cancer effect against blood cancer, breast cancer and neuroblastoma in immune-mediated manner. RBO displays a dose dependent amelioration of myelo-suppression in cyclo-phosphamide-treated rats (Park et al, 2013, 2017). Hence, various components of rice bran are effective in stimulating the immune system against various diseased conditions.

**Hepato-protective effects**

Liver injury is directly related to the activity of plasma...
aspartate aminotransferase, alanine aminotransferase and hepatic lipid hydroperoxide levels. To reduce the liver injury, these components should be reduced. Chotimarkorn and Uschio (2008) studied the outcomes of γ-oryzanol and ferulic acid on liver damage and observed that oral administrations of these compounds display a defence mechanism on liver injury, which is originated ingestion of chronic ethanol ingestion. This is attributed towards the inhibition of liver damaging enzymes i.e. plasma alanine aminotransferase and aspartate aminotransferase by ferulic acid and γ-oryzanol present in rice bran. Increased activity of superoxide dismutase assists in overcoming the liver injury of the mice when treated with ferulic acid and γ-oryzanol (Park et al, 2014).

**Applications of RBO**

Bioactive compounds such as tocopherols, tocotrienols, γ-oryzanol and unsaturated fatty acids make RBO an excellent candidate to enrich foods. RBO provides many health promoting functions and acts as an antioxidant, and increases mass of muscles and reduces oxidized and cholesterol serum cholesterol levels (Park et al, 2017; Bumrungpert et al, 2019). To increase oxidative stability and to enhance nutritional value, RBO is added to the food systems.

**Food applications**

**Cooking oil**

Most of the oil seeds have strong flavours which cover the natural flavour of food. RBO has mild flavour and pleasant smell, therefore, can be used as an essential ingredient in cuisines around the world. High smoke point of 232 °C and ignition point of 350 °C make RBO fit for high-temperature cooking methods (Lai et al, 2019). RBO is produced sustainably with hypoallergenic profile and can be used as a substitute for traditional cooking oil for those who are allergic (Nayik et al, 2015). RBO also assists in calming down the hyper sensitivity to other allergens and also stops allergic reaction in the human body. RBO is rich in antioxidants and protects hairs from UV exposure, further makes it popular as cooking oil in households. As compared to other vegetable oils, peroxide value, acid value, degree of polymerization and total polar compounds of RBO were demonstrated by a slow degree of increase upon deep frying (Nayik et al, 2015). Among various oils, RBO possesses the highest viscosity because of appreciable amount of oryzanol content (> 10 g/kg) because of its good dressing performance in Chinese cuisine. High viscous behaviour of RBO makes the food shiny and appetizing by retaining on food surface (Wang, 2019). High smoke point, ignition point, viscosity, hypoallergenic profile and other beneficial effects compose RBO as perfect oil for cooking purposes.

**Bakery products**

Lipids impart mouthfeel, lubrication, tenderization, air incorporation structural integrity, and shelf life to the bakery goods. Due to lipid’s creaming ability, plastic shortenings, which are normally produced by a partial hydrogenation process, are commonly used in the baking industry (Zhou et al, 2011). During the hydrogenation process, trans-fatty is produced and increases the risk of coronary diseases. Further, RBO has more micro-nutrients, flavour and taste. Frying is efficient which makes RBO more economical (15% less absorption during frying) and also saves energy. As per FDA and USDA recommendations, saturated fats should be replaced with MUFA/PUFA, and intake of trans-fat should also be reduced so as to prevent the risk of heart ailments. A number of approaches have been practiced to minimize trans-fatty consumption either by blending of oils with high oleic and low linolenic fatty acid with fully hardened oils or by randomizing through interesterification (Jeyarani et al, 2009). RBO may be used in baked foods as a fat replacer with high oxidative stability. Shaik et al (2017) replaced hydrogenated fat with RBO, and two types of RBO spread (RBOS1 and RBOS2) are used for the preparation of cake, and found that the major trans-fatty, i.e. elaidic acid and linolelidic acid are the highest in hydrogenated fat cake and the least in RBO cake. Therefore, excellent frying properties, high oxidative stability and fatty acid profile of RBO make it a feasible option for the food manufacturers for the development of the zero or low trans-fatty products.

**Milk and milk products**

Moisture uptake and auto-oxidation are major causes of deterioration during processing and storage in whole milk powder (WMP). Auto-oxidation in WMP is due to the presence of polyunsaturated monocarboxylic acids in milk fat. Oxidation of milk fats occurs through chain reaction of free radicals, and the antioxidants participate in the reaction and stabilize these free radicals to prevent deterioration of the milk (Nanua et al, 2000). The addition of 0.1% RBO significantly improves the oxidation stability of low-heat WMP. This effect is attributed to the antioxidants
present in RBO. Food grade antioxidants are mono- or poly-hydric phenols with various ring substitutions. Milk contains some phytonutrients, vitamins (vitamins C and E) and β-carotene, however, their concentrations depend on their abundance in the feed (Elliott, 1999). Many attempts have been made to enhance milk’s vitamin E content by supplementing it in feed but only 2% of the ingested vitamins are transferred to milk. RBO is a rich source of vitamin E (tocopherols and tocotrienols), which has well-studied antioxidative properties. Nanua et al (2000) incorporated 0.1% and 0.2% RBO in milk and indicated that addition of RBO is effective in reducing the oxidation of WMP without imparting a detectable flavour change. Abbas et al (2017) fortified yoghurt by the incorporation of 3% RBO and Bifidobacterium strain, and found that yoghurt has the satisfactory viscosity as well as potential health benefit of RBO. RBO was incorporated into soft cheeses to study its oxidative stability, microbial and sensory properties (El-Galeel and Atwaa, 2017). The results showed that the investigated cheese shows high content of phenolic components and exhibits a good oxidative stability, with better sensory properties and the lowest microbial count than the butylated hydroxy anisole treatments. The antioxidants and an exceptional compatibility with milk based components make RBO a suitable ingredient for development of innovative products.

**Meat and meat products**

Sausages are formulated by adding 30% of animal fat to impart emulsion and gelling characteristics to meat products. Consumption of high animal fat is associated with increased risk of obesity, CVDs and hypertension. Efforts have been carried out to formulate sausages with low fat/calorie, and phosphate free by replacing animal fat with plant oil (Yum et al, 2018). The increased demand for RBO as a functional food ingredient has led to an increase of defatted rice bran supply obtained after oil extraction. RBO is a natural ingredient that meets consumers demand for healthy meat products. Moreover, RBO addition to meat products increases the value for a secondary by-product from rice-processing plants (da Veiga et al, 2020). The pork sausage prepared by substituting pork back fat with 35%, 40%, 45% and 50% RBO. The sensory and physicochemical properties of the innovative sausage were compared with the control sausage. RBO showed the lowest cooking loss (40%) compared to the sausage prepared from pork back fat. RBO added to sausages not only improves the tenderness and softer texture but significantly increases the UFA and PUFAs. Further, RBO substitution has not imparted any sensory or flavor to the developed sausage. The neutral taste, antioxidants and improved physicochemical properties of RBO make it a perfect option for substitution of animal fats.

**Structured lipids (SLs)**

SLs are basically TAGs which are modified by incorporating new fatty acids and/or to change their place distribution in glycerol backbone. MCFAs are transported and metabolized through the portal bloodstream rather than lymphatic system, and help in providing quick energy sources which is oxidized and utilized for fuel. MCFAs are often incorporated into TAGs to produce SLs (Jennings et al, 2010). They prepared RBO SLs using RBO and caprylic acid by utilizing lipozyme RM IM (immobilized lipase on a macroporous anion exchange resin), as a biocatalyst. The increased interest in SLs may be attributed to its low caloric intake and no harmful effects associated with the ingestion of medium-long-medium lipids.

**Emulsifier**

Rice bran lecithin (RBL), a by-product of RBO, is a mixture of phospholipids and a novel choice as an emulsifier for food industry. RBL has potential to form nanoemulsions due to its emulsifying properties (Lehri et al, 2020). Sun et al (2020) recovered RBL from enzymatic de-gumming catalyzed by phospholipase A1 and reported it with better emulsifying properties compared with RBL from citric acid degumming and water degumming, which may be useful for industrial application.

**Non-food applications**

**Polymers**

Biodegradable polymers (bioplastics) are degradable, easily de-polymerized and offers an interesting alternative to petroleum derived polymers. Among all, poly(lactic acid) (PLA) is widely used for packaging purposes. To enhance the ductility of PLA materials, different strategies have been adopted i.e. polymer blending, copolymerization as polymer additives to improve its flexibility and workability, to maintain the integrity of film and to avoid pores and cleavages. Preparation of RBO-based natural plasticizer has interesting characteristics compared to commercially used plasticizer. Vieira et al (2014) synthesized RBO-based natural plasticizer by esterification reaction of RBO fatty acid and polyols, and investigated its preliminary application in polyvinyl chloride (PVC).
The results indicated that incorporating RBO plasticizer to PVC films significantly increases the elongation at break (371.2%) compared to pure PVC film. The high plasticizing properties of RBO-based PVC films were due to high flash point of RBO. RBO added to PLA as plasticizer in small concentration and found useful for a faster preparation of PLA materials by accelerating the growth of PLA α-crystals at a low crystallization temperature (Righetti et al, 2019). The good compatibility with other resins, low toxicity and chemical structure of RBO make suitable for developing the polymeric material with superior properties.

**Lubricant and bio-fuel**

The poor biodegradability of petroleum oils pressurizes the industry to develop eco-friendly biodegradable lubricants from agricultural feed stocks. The bio-lubricants are given greater consideration as renewable and potential source of energy in future especially in the developing countries (Rani et al, 2015). The biodegradability, renewability, low toxicity and excellent lubricating performance of vegetable oils are the main reason behind the use of vegetable oil in the upcoming bio lubricant formulations. The vegetable oil generally has high flash point, fire point, high viscosity index, high lubricity and low evaporative losses. The physiochemical and thermal properties of RBO are good compared to other vegetable oils because of good thermal and tribological properties. RBO has good frictional properties compared to SAE20W40 (commercially available mineral oil), but the wear scar diameter is more for RBO and can be improved by adding proper anti-wear additives.

The depletion of non-renewable energy sources, especially diesel, petroleum and fossil fuel, has warned the global community switch to energy alternatives. Being non-toxic, biodegradable, and renewable, biodiesel received increased attention and RBO is a good option as raw material for biodiesel production. During the process of biodiesel production from RBO, carbohydrates, proteins and purified nutraceuticals are also generated, which makes the biodiesel inexpensive. Production of biodiesel is advocated either via in situ esterification, lipase-catalyzed esterification, and acid-catalyzed or base-catalyzed reactions (Ju and Vali, 2005).

**Cosmetics**

Increasing demand for natural and value-added healthy ingredients grows RBO as an important ingredient to the personal care and cosmetics industry. RBO nano-emulsions improve skin’s moisture and physical stability, therefore may be used in anti-ageing and sunscreen formulations cosmetics (Bernardi et al, 2011; Mukhopadhyay and Siebenmorgen, 2017). Vitamin E present in RBO nourishes skin cells and preventing pore clogging due to skin regenerates and slows down the aging process (Nayik et al, 2015). RBO based nanoemulsion provide skin protection against UV and Infrared radiation and skin diseases, i.e. atopic dermatitis and psoriasis. Sari et al (2020) prepared oil-in-water nanoemulsions using coconut or palm oil combined with RBO, and reported that RBO containing nanoemulsion has good stability with zeta potential values of < -30 mV. Rice bran extracts and RBO have protective action against skin allergies/diseases, injuries against UV-B radiations and also exerts anti-aging properties. Due to various beneficial effects, RBO can be used as bath, moisturiser and numerous other cosmetic products.

**CONCLUSIONS**

RBO is a balanced source of SFA, MUFA and PUFA along with a rich source of γ-oryzanol, tocopherols, tocotrienols and better oxidative stability than other cooking oils. This review highlighted the latest findings on the extraction and functionalization of the RBO for food and non-food application. Their utilization is in direct alignment with their functional and structural properties, which can be directly altered by their extraction procedures. Recent scientific development has established RBO extraction techniques with respect to growing demands for process improvement with special consideration to the operational speed, repeatability and predictability of the products. As underlined above, various novel and efficacious method are being validated for extraction of the highest quality of RBO. The nutritional composition, biological activities and health benefits of RBO have attracted the attention of consumers for its wider application. The constituents of RBO have also gained its application in numerous non-food and food applications. The future focus of many research organizations should be the identification and use of novel extraction technologies including hybrid techniques for RBO extraction. Integrating RBO, nanomaterials, fatty acids, phenolic compounds and other natural bioactives can assist the development of functional biocomposites, which might have emerging application in the food packaging system.

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