

# Chemopreventive Properties of Dietary Rice Bran: Current Status and Future Prospects<sup>1,2</sup>

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## ABSTRACT

Emerging evidence suggests that dietary rice bran may exert beneficial effects against several types of cancer, such as breast, lung, liver, and colorectal cancer. The chemopreventive potential has been related to the bioactive phytochemicals present in the bran portion of the rice such as ferulic acid, tricin,  $\beta$ -sitosterol,  $\gamma$ -oryzanol, tocotrienols/tocopherols, and phytic acid. Studies have shown that the anticancer effects of the rice bran-derived bioactive components are mediated through their ability to induce apoptosis, inhibit cell proliferation, and alter cell cycle progression in malignant cells. Rice bran bioactive components protect against tissue damage through the scavenging of free radicals and the blocking of chronic inflammatory responses. Rice bran phytochemicals have also been shown to activate anticancer immune responses as well as affecting the colonic tumor microenvironment in favor of enhanced colorectal cancer chemoprevention. This is accomplished through the modulation of gut microflora communities and the regulation of carcinogen-metabolizing enzymes. In addition, the low cost of rice production and the accessibility of rice bran make it an appealing candidate for global dietary chemoprevention. Therefore, the establishment of dietary rice bran as a practical food-derived chemopreventive agent has the potential to have a significant impact on cancer prevention for the global population. *Adv. Nutr.* 3: 643–653, 2012.

## Introduction

Despite major advances in research, cancer remains one of the main causes of mortality worldwide (1,2). The WHO has estimated that in accordance with current trends, the global cancer burden will continue to increase, with 70% of new cases predicted to occur in the developing world (1). As a result, there is a great need for the development and implementation of novel, effective, and affordable chemopreventive strategies that can reach a diverse global population. In developed countries, chemopreventive drugs, such as nonsteroidal anti-inflammatory drugs and tamoxifen, are being used in high-risk populations; however, due to safety risks and high cost, these drugs are not feasible in the developing world (3–5). On the other hand, consumption of certain whole foods (i.e., grains, fruits, vegetables, teas, and spices) has been associated with reduced cancer

risk, and their inherent safety makes these functional foods an appealing choice for widespread, long-term use in diverse populations (6). In further support of this dietary chemopreventive angle, the American Association for Cancer Research and the World Cancer Research Fund has estimated that 30–40% of all cancers can be prevented by appropriate diet and body weight (7). As a result, there has been increased interest in the identification and characterization of food components with potential chemopreventive properties—most importantly, foods with the capacity to be grown in many regions of the world.

Epidemiological studies suggest that the fiber from whole grains is significantly associated with a lowered risk of colorectal cancer (CRC)<sup>6</sup> (8–10). Rice, *Oryza sativa*, is a staple food crop for more than half of the world's population (11). Although brown rice is a whole grain, most of the world, including the United States, consumes white rice (12). As a result, research attention has been directed toward

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<sup>6</sup> Abbreviations used: AOM, azoxymethane; CF, cycloartenyl ferulate; COX, cyclooxygenase; CRC, colorectal cancer; DC, dendritic cell; iNOS, inducible nitric oxide synthase; NF- $\kappa$ B, nuclear factor- $\kappa$ B; NK, natural killer; PGE<sub>2</sub>, prostaglandin E<sub>2</sub>.

understanding the chemopreventive properties associated with brown rice and rice bran. The bran fraction of rice is composed of phytochemicals and nutrients with known cancer-fighting and immune-enhancing properties (4). Furthermore, various international rice cultivars exhibit phytochemical diversity that may translate into differential anticancer effects (13). Comparative studies on the chemopreventive properties of rice bran across diverse varieties are still needed. There are review articles on the chemopreventive potential of fiber and other whole grain components (7–10,14), yet there has not been, to our knowledge, a comprehensive review focusing on the chemopreventive properties of rice bran. Therefore, the major objective of this review is to provide a cohesive representation of the literature on the effects and mechanisms for dietary rice bran cancer chemoprevention.

### Rice bran and brown rice phytochemicals

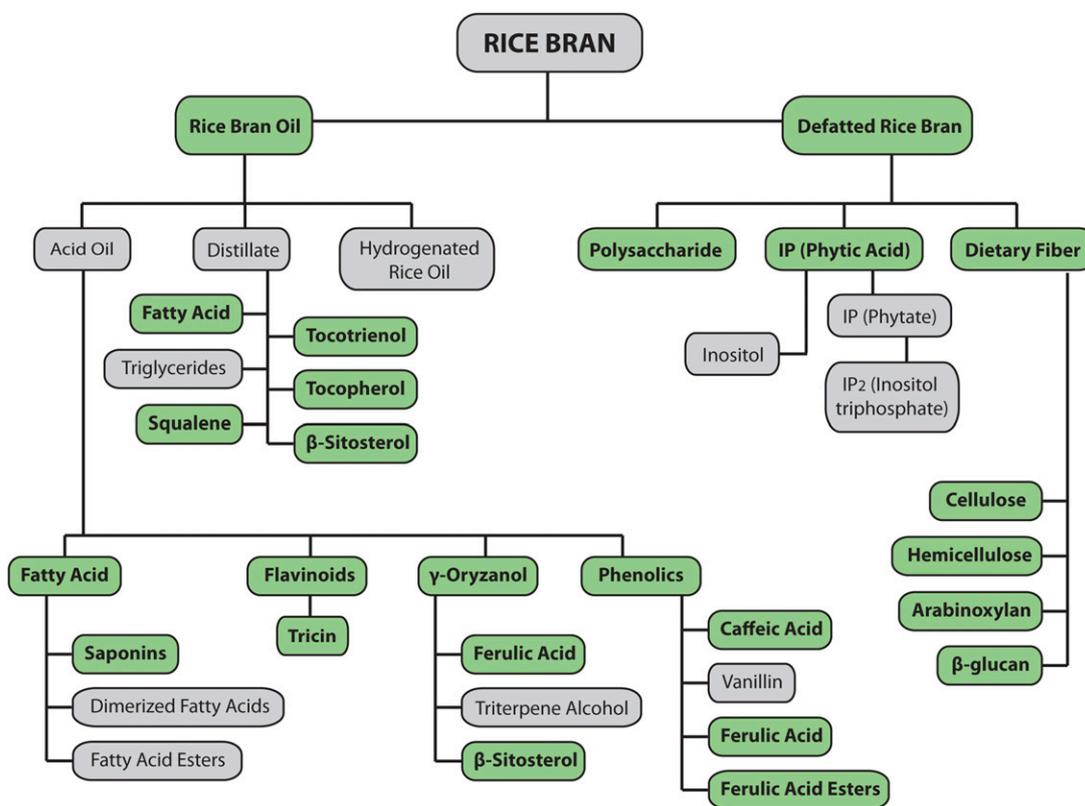
Rice bran contains a variety of bioactive components with chemopreventive activity, including  $\gamma$ -oryzanol, ferulic acid, caffeic acid, triclin, coumaric acid, phytic acid; the vitamin E isoforms  $\alpha$ -tocopherol,  $\gamma$ -tocopherol, and various tocotrienols; phytosterols such as  $\beta$ -sitosterol, stigmasterol, and campesterol; and carotenoids (15) such as  $\alpha$ -carotene,  $\beta$ -carotene, lutein, and lycopene (Fig. 1). Rice bran also contains cellulose, hemicellulose, pectin, arabinoxytan, lignin, and  $\beta$ -glucan; micronutrients such as calcium, magnesium, and 9 B vitamins; and essential amino acids such as

tryptophan, histidine, cysteine, and arginine (16). Compared with other cereal brans, such as corn, wheat, and oat, the lipid fraction of rice bran contains a unique ratio of vitamin E isoforms ( $\alpha$ -,  $\gamma$ -,  $\delta$ - tocotrienols and tocopherols),  $\gamma$ -oryzanol, and  $\beta$ -sitosterol. Previous studies in our laboratory and others further suggest that the process of fermenting rice bran with bacterial or fungal agents can beneficially alter the bioactivity (17–20). For example, fermenting rice bran with *Saccharomyces boulardii* induced an increase in the amount of ferulic acid released and reduced lymphoma cell viability compared with nonfermented rice bran (18). Many plant phenols, such as ferulic acid, are often biologically unavailable after intake, and fermentation helps improve the efficacy of the antioxidant activities (21). Fermented brown rice demonstrated cancer-fighting properties in vivo (Table 1) and merits continued investigation of the changes in bioavailability, metabolism, and anticancer activities after human consumption.

### In vivo cancer chemopreventive studies

Numerous animal studies have been performed to investigate the cancer-fighting properties of dietary rice bran across tumor types. The cancer chemopreventive and control mechanisms attributed to dietary rice bran intake are summarized in Table 1.

**Animal studies.** The effects of dietary rice bran intake have been extensively studied in the azoxymethane (AOM)



**Figure 1.** A breakdown of the components found in dietary rice bran. Green boxes represent components with literature-supported cancer chemopreventive activity.

**Table 1.** In vivo studies highlighting the chemopreventive nature of dietary rice components<sup>1</sup>

Cancer type	Species	Rice component	Mechanism of action	Ref
Colon/gastric	Fisher 344 rats	Rice bran	Decreased the incidence of large bowel tumors	(95)
	Apc <sup>Min</sup> mice	Rice bran (30%)	Decreased numbers of intestinal adenomas and decreased intestinal hemorrhage	(4)
	Fisher 344 rats	Rice bran hemicellulose	Decreased the incidence and total number of colon tumors	(92)
	Fisher 344 rats	Rice bran oil	Decreased the incidence of aberrant crypt foci formation	(24)
	Fisher 344 rats	Ferulic acid and defatted rice germ	Decreased the number of aberrant crypt foci	(23)
	Apc <sup>Min</sup> mice	Tricin from rice bran	Decreased the number of intestinal adenomas potentially via the inhibition of COX enzyme activity and the decrease in PGE <sub>2</sub> production	(26)
	Sprague-Dawley rats	Phytic acid extracted from rice bran	Decreased the number of aberrant crypt foci	(25)
	ACI rats	Fermented brown rice by <i>Aspergillus oryzae</i>	Decreased the total number of gastric lesions	(27)
	Apc <sup>Min</sup> mice	Fermented brown rice and bran	Suppressed the multiplicity of colon tumors in DSS-exposed mice through the induction of anti-inflammatory activity in the colon	(17)
	Fisher 344 rats	Fermented brown rice and bran	Inhibited aberrant crypt foci formation, suppressed the incidence and multiplicity of colon adenocarcinomas, and inhibited cell proliferation	(22)
Humans	Brown rice	Consumption of brown rice at least once weekly reduced the risk of colorectal polyp formation by 40%	(37)	
Breast	MF-1 nude mice	Tricin	Decrease in the volume of tumors after the implantation of MDA-MB-468 cells pretreated with tricin	(34)
	Sprague-Dawley rats	Tocotrienol-rich fraction of rice bran oil	Decreased the severity and number of tumors in the mammary glands	(33)
	BALB/c mice	Sulfated polysaccharide (SRBPS2a) from defatted rice bran	Inhibited the growth of implanted EMT-6 tumor cells	(35)
Lung	A/J mice	Fermented brown rice and bran	Decreased size and multiplicity of tumor, which is possibly due to the induction of CyP2a5 and the resulting decrease in tumor cell proliferation	(32)
Oral	Fisher 344 rats	Fermented brown rice by <i>Aspergillus oryzae</i>	Decreased the incidence and multiplicity of tongue carcinoma	(31)
	Fisher 344 rats	Ferulic acid and defatted rice germ	Decreased the incidence of tongue carcinomas and preneoplastic lesions	(23)
Bladder	ICR mice	Fermented brown rice by <i>Aspergillus oryzae</i>	Decreased the incidence of urinary bladder carcinoma and preneoplastic lesion formation	(30)
Liver	Sprague-Dawley rats	Tocotrienol-rich fraction from rice bran oil	Decreased the risk of hepatocarcinoma via decreased nodule formation in the liver. Also resulted in decreased plasma alkaline phosphatase levels, decreased activity in liver glutathione transferase, and decreased LDL and lipid peroxidation	(33)
	F344 rats	Fermented brown rice and bran	Decreased the incidence and multiplicity of hepatocellular carcinoma during the pre- and postinitiation phases	(28)
Esophageal	F344 rats	Fermented brown rice by <i>Aspergillus oryzae</i>	Suppressed the total number of esophageal tumors and dysplastic lesions potentially via the inhibition of cell proliferation	(29)
Melanoma/skin	C57/Bl6 mice	Exo-biopolymer from rice bran cultured with <i>Lentinus edodes</i>	Suppressed the growth of the transplanted B16/B16 melanoma tumor after oral rice bran treatment potentially via the activation of NK cells	(36)
	ICR mice	Cycloartenol ferulate extracted from rice bran	Inhibited tumor promotion in 2-stage skin carcinoma potentially via blocking inflammation	(52)

<sup>1</sup> COX, cyclooxygenase; DSS, DSS, dextran sodium sulfate; NK, natural killer; PCE<sub>2</sub>, prostaglandin E<sub>2</sub>.

carcinogen-induced CRC model. Aberrant crypt foci formation is one of the earliest observable changes associated with AOM-induced CRC development. In these studies, a decrease in the total number of aberrant crypt foci was observed in mice consuming diets supplemented with a wide variety of rice bran components, including regular and defatted rice germ, phytic acid extracted from rice bran, and rice bran

oil (22–25). The study by Panala et al. (24) revealed decreased tumor incidence in rice bran oil diet–fed AOM-treated Fisher 344 rats compared with the control-fed animals after 45 wk of dietary intervention. Furthermore, the suppression of AOM-induced tumor incidence and multiplicity was also observed after consumption of fermented brown rice. These authors concluded that one of the chemopreventive mechanisms of

the fermented brown rice was inhibition of the cellular proliferation of the colonic mucosa (22).

Dietary rice bran has also been studied for chemopreventive effects in the *APC*<sup>min/+</sup> mouse model for intestinal carcinogenesis, which resembles the human heritable condition familial adenomatous polyposis. This transgenic model has been valuable for displaying similar gene expression profiles, inflammation, and chemotherapeutic responses, as seen in humans. A study by Verschoyle et al. (4) showed a reduction in the number of tumors in mice consuming a 30% rice bran diet and identified the fibrous part of the bran to be responsible for the chemoprevention. Another group showed reduced intestinal adenoma numbers as a result of tricetin, a unique rice bran-extracted flavone. These authors postulated that the reduced tumor numbers were due to tricetin inhibiting the cyclooxygenase (COX) enzymes in a similar fashion as nonsteroidal anti-inflammatory drugs. This finding was confirmed and demonstrated through reduced prostaglandin E (PGE<sub>2</sub>) levels in the serum and the intestinal mucosa of mice fed the tricetin-rich diet (26). In another study, fermented brown rice was also shown to prevent inflammation-induced intestinal carcinogenesis in *APC*<sup>min/+</sup> mice treated with dextran sodium sulfate. Mice that were fed the fermented rice bran diet had decreased total numbers of tumors per mouse compared with control diet-fed mice. The decrease in cyclooxygenase 2 (COX-2) and inducible nitric oxide synthase (iNOS) mRNA expressed in the colonic mucosa led them to conclude that rice bran induced an anti-inflammatory environment that helped to protect against tumorigenesis (17).

The effects of fermented brown rice on the development of gastric cancer have also been investigated. Mice fed a diet containing 5% or 10% fermented brown rice for 52 wk showed a decreased incidence and multiplicity of gastric tumors compared with control diet-fed mice. The authors suggest that the ability of the fermented brown rice to block cell proliferation may be responsible for the observed gastric cancer chemoprevention (27).

In addition to colonic and gastric cancers, fermented brown rice has demonstrated chemopreventive effects against cancers of the lung, liver, bladder, tongue, and esophagus (28–32). All of these studies were able to demonstrate that the feeding of fermented brown rice to carcinogen-treated mice significantly decreased the incidence and multiplicity of the tumors in the respective tissues. Furthermore, the study by Phutthaphadoong et al. (32) revealed the ability of fermented brown rice to inhibit 4-(methylnitrosamino)-1-(3-pyridyl)-1-butanone-induced lung carcinogenesis that is associated with decreased Cyp2a5 mRNA expression levels. Cyp2a5 is a metabolic activator known to enzymatically cleave inactive carcinogens. Also, in the studies focused on carcinogenesis of the tongue and esophagus, fermented brown rice-fed mice were shown to have decreased incorporation of bromodeoxyuridine in the non-lesion esophageal and oral squamous epithelial tissues compared with control diet-fed mice (29,31). These authors postulated that the ability of the fermented brown rice diet

to inhibit the carcinogen-induced hyperproliferation of the target organ tissue was an important mechanism of its cancer chemopreventive activity.

Several dietary rice bran components have also shown cancer chemopreventive activity against breast cancer. A study by Iqbal et al. (33) revealed the capacity of a diet rich in the tocotrienol fraction of rice bran oil to protect Sprague-Dawley rats against the mammary carcinogen 7,12-dimethylbenz[ $\alpha$ ]anthracene. Tocotrienol diet-fed animals exhibited decreased neoplastic transformations that were in part due to the decreased alkaline phosphate levels in the serum and mammary tissues and were suggestive of reduced carcinogen-induced tissue damage. Additionally, it has been shown that the bioactive components, tricetin and a sulfated polysaccharide, extracted from rice bran, were capable of inhibiting the tumor growth and volume in mouse models of implanted human-derived breast cancer cells (34,35). The distribution of tricetin in the blood and tissues after consumption of the 0.2% tricetin-rich diet was not sufficient to affect the tumor burden, and the authors further investigated the direct effects of tricetin incubation with tumor cells before implantation into mice. This study cautions that before tricetin can be considered a promising candidate for breast cancer chemoprevention, optimization studies in humans are required to determine bioavailability and absorption (34).

Fermented rice bran extract has also demonstrated cancer chemopreventive activity through the activation of specialized immune cells. A study by Kim et al. showed that an exo-biopolymer extracted from fermented rice bran has the ability to suppress the growth of a transplanted melanoma tumor through the activation of natural killer (NK) cells. This study suggests that after oral administration of the fermented rice bran extract, a variety of rice bran-derived bioactive components aid in stimulating the gut-associated lymphoid tissue as a means for mediating cancer chemoprevention in mice (36).

**Human studies.** Rice is a staple crop for more than half of the world's population and accounts for >20% of global energy intake (11). White rice is consumed in higher amounts compared with the whole-grain brown rice that includes the bran. One of the major barriers to whole-grain brown rice or rice bran availability for consumption is the short shelf life due to high lipid content of the bran. Heat stabilization or fermentation are practical means to prolong the shelf life for increased consumption globally. In 2011, Tantamango et al. (37) reported a prospective study that revealed an association between the consumption of different foods and the risk of the development of rectal/colon polyps. In addition to protection seen after the consumption of green vegetables, dried fruit, and legumes, this study found that consumption of brown rice had the strongest correlation with cancer chemoprevention. They showed that the consumption of brown rice at least once per week was associated with a 40% reduced risk of polyp formation. The authors concluded that this correlation may be related to

the high fiber content of brown rice, the phytates, and/or other bioactive components that have been hypothesized for exhibiting chemoprevention efficacy.

Numerous studies have shown MGN-3 (a fungal fermented rice bran product) to have anticancer activity (38,39) and to modulate the immune response (38,40–43). MGN-3 has also been shown to be a potent activator of NK-cell activity in cancer patients as well as an inhibitor of T-regulatory cells responsible for cancer-related immunosuppression (44). Statistical data on accurate human consumption of fermented brown rice or fermented rice bran products such as red rice yeast are limited. The Food and Agriculture Organization reports 215,000 tons of fermented brown rice was consumed in Japan (45). Fermented foods, in general, are receiving increased attention in food science research because of the proven safety and potential abilities to prevent and/or treat chronic diseases (46).

### In vitro cancer chemopreventive studies

In addition to the investigation of dietary brown rice and rice bran efficacy in animal models of carcinogenesis, a substantial amount of work has been performed in vitro that advances our knowledge of mechanisms involved in the cancer chemopreventive activity of rice bran components. Dietary rice bran and its bioactive components have been shown to inhibit carcinogenesis by inhibiting proliferation, inducing apoptosis, suppressing chronic inflammation, scavenging free radicals, blocking cell-signaling pathways, and activating enzymes known to detoxify carcinogens (Table 2). Whole rice bran extracts and purified single components may also alter cell cycle progression in cancer cell lines. No adverse effects have been observed in response to the rice bran components on normal cells, which provides important evidence of the intrinsic safety of dietary rice bran in cancer chemoprevention (18,47,48).

**Table 2.** In vitro studies highlighting the chemopreventive nature of dietary rice bran extracts

Cancer type	Cell line(s) <sup>1</sup>	Rice bran component(s)	Mechanism of action	Ref
Colon/gastric	SW480 and SW620	Cycloartenyl ferulate	Induced apoptosis via enhanced activation of caspase-8 and -3	(48)
	SGC-7901	$\gamma$ -Tocotrienol	Inhibited cell growth via cell cycle arrest and induction of apoptosis by activation of caspase-9 and -3	(49)
	SW480, HT-29, and HCEC	Phenolic extracts (tricin, ferulic acid, caffeic acid, and methoxycinnamic acid)	Decreased the number of viable SW480 and HCEC cells and reduced the colony-forming ability of these cells	(51)
	Caco-2	Peptide hydrolysates derived from defatted rice bran	Inhibited cell proliferation	(96)
	Caco-2 and HCT-116	Pentapeptide derived from defatted rice bran	Inhibited cell growth	(53)
	Breast	EMT-6 (murine)	Sulfated polysaccharide (SRBPS2a) from defatted rice bran	Inhibited cell growth
MDA-MB-468		Tricin	Inhibited cell growth via cell cycle arrest within G <sub>2</sub> /M phase	(34)
MDA-MB-468, MCF-7, and HBL100		Phenolic extracts (tricin, ferulic acid, caffeic acid, and methoxycinnamic acid)	Decreased the numbers of viable MDA-MB-468 and HBL100 cells and reduced the colony-forming ability of these cells	(51)
MCF-7, ZR75-1, and HCC-70		Denatured rice bran hemicellulose (MGN-3/Biobran)	Enhanced yeast-mediated apoptosis via activation of caspase-8, -9, and -3	(19)
MCF-7 and MDA-MB-231		Pentapeptide derived from defatted rice bran	Inhibited cell growth	(53)
Liver		HepG2	Peptide hydrolysates derived from defatted rice bran	Inhibited cell proliferation
	HepG2	Pentapeptide derived from defatted rice bran	Inhibited cell growth	(53)
Mesothelioma	H28	$\gamma$ -Tocotrienol	Decreased cell viability and increased cytotoxicity via inhibition of phosphatidylinositol 3-kinase-AKT signaling	(50)
Melanoma	U266	Denatured rice bran hemicellulose (MGN-3/Biobran)	Inhibited cell proliferation by blocking cells from entering G <sub>0</sub> -G <sub>1</sub> phase.	(20)
Monoblastic leukemia	U937	Rice bran agglutinin	Inhibited cell growth via the induction of apoptosis and cell cycle arrest within the G <sub>2</sub> /M phase	(97)
B-cell lymphoma	Raji	<i>Saccharomyces boulardii</i> fermented and nonfermented rice bran extracts	Reduced cell viability	(18)
Endometrial adenocarcinoma	Sawano	Lipoprotein fraction extracted from rice bran	Induced apoptosis and inhibited cell proliferation	(47)

<sup>1</sup> All studies were performed on human cancer cell lines except when otherwise stated. DSS, dextran sodium sulfate

Rice bran-derived components demonstrating cancer chemopreventive activity *in vivo* have been examined for mechanisms of cancer-fighting activity using cancer cell lines. For example, a study by Sun et al. (49) demonstrated the ability of  $\gamma$ -tocotrienol to induce apoptosis in the SGC-7901 gastric cancer cells through the activation of caspase-3 as well as to elicit cell cycle arrest at the  $G_0/G_1$  phase. Another group showed a synergistic effect of the rice bran-derived  $\gamma$ -tocotrienol in combination with the chemotherapeutic agent cisplatin on the viability of H28 mesothelioma cancer cells. These authors demonstrated that the cytotoxic effect was in part due to the inhibition of phosphatidylinositol 3-kinase-AKT signaling by  $\gamma$ -tocotrienol (50). These results are highly significant as several members of the phosphatidylinositol 3-kinase signaling pathway are commonly overexpressed in different types of cancer to favor uncontrolled proliferation and enhanced cell survival.

Several phenolics (e.g., caffeic acid, triclin, ferulic acid, and methoxycinnamic acid) extracted from brown rice have also revealed cytotoxic and anticlonogenic properties when incubated with a number of breast cancer and CRC cell lines. Tricin was shown to be the most potent compound for inhibiting the growth of human SW480 CRC and MDA MB 468 breast cancer cells (51). Another study identified the ability of purified triclin to arrest the growth of MDA MB 468 breast cancer cells in the  $G_2/M$  phase. These authors hypothesized that triclin is able to modulate important protein regulators of the cell cycle pathway, highlighting its potential as a cancer chemopreventive and chemotherapeutic agent (34).

Cycloartenyl ferulate (CF), a phenolic compound found in rice bran oil, has also been identified as a promising cancer chemopreventive agent based on a study demonstrating the inhibition of skin carcinogenesis in mice after topical application of CF (52). Kong et al. (48) investigated the mechanisms associated with CF-induced cancer chemoprevention *in vitro* using the SW480 CRC cell line. These studies revealed the ability of CF to induce apoptosis in SW480 cells via the activation of caspase-8 and -10 of the extrinsic pathway and caspase-9 of the intrinsic pathway. In addition, CF was shown to support the growth and survival of normal colon CCD-18-Co cells.

Wang et al. (35) investigated the novel sulfated polysaccharide SRBPS2a, derived from defatted rice bran, for cancer chemopreventive activity against EMT-6 breast cancer cells. The authors were able to show the ability of SRBPS2a to inhibit the cellular proliferation of EMT-6 cells through the induction of apoptosis. Also, a unique heat-stabilized, gastrointestinal-resistant pentapeptide isolated from defatted rice bran, has been shown to inhibit the growth of cancer cells isolated from the colon, breast, lung, and liver (53). The authors advocated that this food-derived peptide represents an excellent source of protein as well as a less expensive, natural alternative to synthetic cancer chemopreventive agents.

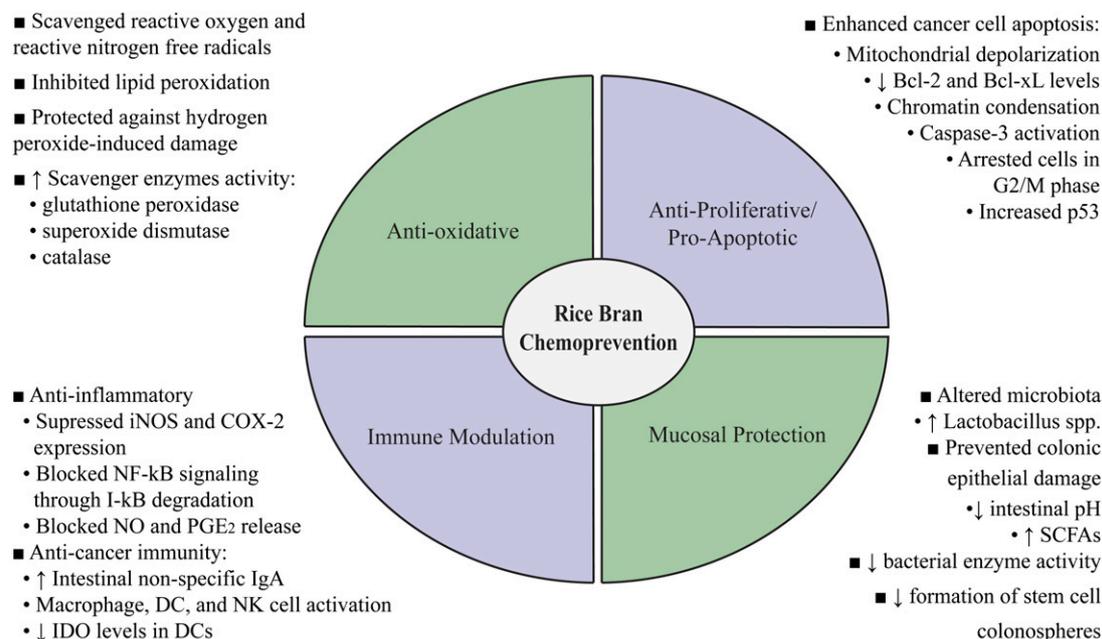
### Rice bran associated cancer chemopreventive mechanisms

There are many stages of the carcinogenic process in which rice bran-derived chemopreventive agents can act to block

or suppress the cellular progression to malignancy. Additionally, there are advantages of the use of dietary rice bran compared with single-agent chemopreventives. Whole dietary rice bran contains a complex mixture of bioactive components, each with a unique capacity to interact with multiple cellular targets to prevent the development of cancer. The next section of this review examines the cancer chemopreventive characteristics of individual rice bran components for the modulation of cellular signaling pathways, immune responses, and microbial communities (Fig. 2).

**Protection against free radicals.** One effective mechanism for preventing cancer is to block initiation via inhibition of the DNA damage that results from reactive oxygen species or other carcinogens (6). Oxidative stress can cause significant cellular damage and irreversible mutations; therefore, sufficient amounts of dietary antioxidants may help to protect cells from free radical damage. Dietary rice bran is an excellent source of phytochemicals with antioxidative properties, such as  $\beta$ -sitosterol and a wide variety of phenolics and carotenoids. Ferulic acid, a well-studied phenolic compound, has been shown to be an effective scavenger of superoxide anion radicals and inhibitor of lipid peroxidation (54). One study demonstrated that ferulic acid protected against hydrogen peroxide-induced cellular damage through increased cellular levels of heme oxygenase-1 and heat shock protein-70 (55).  $\beta$ -Sitosterol, a unique phytosterol found in rice bran, exhibited cancer chemopreventive potential via its ability to scavenge nitric oxide free radicals as well as attenuate cellular  $\beta$ -catenin and proliferating cell nuclear antigen levels (56). Noaman et al. (39) demonstrated the ability of the rice bran supplement MGN-3 to enhance the activity of the reactive oxygen species scavenger enzymes glutathione peroxidase, superoxide dismutase, and catalase in tumor-bearing mice. The rice bran-derived phytochemicals  $\gamma$ -oryzanol, coumaric acid,  $\beta$ -glucan, and triclin (57–59) have also exhibited antioxidant properties, but are yet to be established as cancer chemopreventive agents. The scavenging of free radicals by rice bran compounds highlights an important aspect of the cancer chemopreventive abilities properties.

**Inhibition of cancer cellular proliferation.** In addition to preventing the initiation stage of cancer, it is equally important for cancer chemopreventive agents to be able to block proliferation and/or induce the apoptosis of malignant cells. Many of the bioactive components identified in dietary rice bran such as ferulic acid (60),  $\gamma$ -oryzanol, phytic acid (61), coumaric acid (62), pectin (63), triclin, and tocotrienol-tocopherols have been shown to inhibit tumor growth and/or induce apoptosis (64). Kannappan et al. (65) showed that  $\gamma$ -tocotrienol has the ability to down-regulate the expression of the antiapoptotic proteins Bcl-2 and Bcl-xL as well as induce the expression of SHP-1, which directly inhibits STAT3 activation, the results being significant in the context that the STAT3 pathway is associated with cancer progression through the suppression of apoptosis. Another group was able to demonstrate the induction of apoptosis in breast



**Figure 2.** Diagram depicting chemopreventive mechanisms of action associated with dietary rice bran and brown rice bioactive components. COX-2, cyclooxygenase-2; DC, dendritic cells; IDO, indoleamine 2,3-dioxygenase; iNOS, inducible nitric oxide synthase; NF-κB, nuclear factor-κB; NK, natural killer; NO, nitric oxide; PGE<sub>2</sub>, prostaglandin E<sub>2</sub>.

cancer cells after the exposure of lipophilic caffeic and ferulic acid derivatives. These phytochemicals were shown to be responsible for the increase in the expression levels of tumor suppressor protein p53 as well as increase in mitochondrial depolarization and chromatin condensation (66). β-Sitosterol has also been shown to exhibit beneficial effects against cancers of the colorectal, stomach, and breast (67, 68). For example, studies have shown the involvement of β-sitosterol in increased cancer cell membrane damage, activation of caspase-3 (68), and increased cellular ceramide production, which is associated with the induction of cell cycle arrest (67). In addition, numerous flavonoids including tricetin, quercetin, and apigenin have been shown to influence the cell cycle machinery and survival pathways in malignant cells (6). A study by Cai et al. (34) revealed the ability of tricetin to arrest breast cancer cells in the G<sub>2</sub>/M phase of the cell cycle through what was hypothesized to be the modulation of cell cycle protein regulators, such as p34<sup>cdc2</sup>, Wee1, and p21<sup>cip1</sup>. In brief, the ability of rice bran-derived bioactive compounds to inhibit cancer cell proliferation seems to be the key mechanism involved in protection against cancer progression.

**Modulation of immunity and inflammation.** A variety of bioactive components found in dietary rice bran have been shown to aid in cancer chemoprevention through either the enhancement or suppression of the immune response (17,36,69). The early stages of an inflammatory response are crucial for protection against infection and injury, but an uncontrolled, chronic inflammatory environment has been shown to favor cancer development. In particular, proinflammatory cytokines such as TNF-α, IL-1, and IL-6 induce nuclear factor-κB (NF-κB) signaling and the expression of iNOS and COX-2.

The overexpression of iNOS and COX-2 can drive tumor development through the accumulation of DNA mutations and the overproduction of prostaglandins (70,71). In this regard, natural compounds such as flavonoids, ferulic acid, coumaric acid, and α-tocopherol, which have the potential to block proinflammatory immune signaling, have been shown to reduce the development of cancer. A ferulic acid derivative has been shown to be a promising cancer chemopreventive agent due to its ability to suppress the LPS-induced protein expression of iNOS and COX-2 as well as to block the release of TNF-α. The authors postulated the phenomenon to be related to the suppression of I-κB degradation and subsequent inhibition of NF-κB signaling, IL-12 production, and Th1 immune responses has also been associated with salicylic acid, a phenolic found in rice bran (73). The anti-inflammatory effects of a variety of flavonoids were revealed to be a result of the ability to inhibit iNOS enzyme expression and subsequent nitric oxide production as well as block the LPS-induced PGE<sub>2</sub> release and COX-2 expression (74). Thus, decreasing inflammation via the inhibition of COX-2 expression and reduction in PGE<sub>2</sub> production is a promising cancer chemopreventive mechanism for dietary rice bran that merits further investigation.

The immune system has a variety of antitumor mechanisms that can be enhanced using bioactive rice bran components. These cancer chemopreventive immune responses act either by eliminating viral infections associated with virus-induced tumors, by inducing prompt resolution of inflammation after infection, or by the specific elimination of tumor cells (75). Antitumor immune responses may involve a transient production of proinflammatory cytokines to activate a cytotoxic immune responses. In this regard, the

important immune effector cells associated with cancer chemoprevention include dendritic cells (DC), macrophages, NK cells, and T and B lymphocytes. Our recent report of whole dietary rice bran modulation of the mucosal immune response, namely, the increase in mesenteric lymph node and lamina propria DC numbers (76) merits further investigation as a mechanism for cancer control and prevention. Studies have revealed the ability of  $\alpha$ -tocopherol to enhance cytokine production and monocyte recruitment, both of which are necessary for the development of adaptive immunity (77). In this regard,  $\alpha$ -tocopherol has been shown to be a necessary component of the ASO3 vaccine adjuvant. In addition, the enhancement of the respiratory burst and interferon- $\gamma$  production in macrophages has been observed in response to  $\alpha$ -tocopherol and ferulic acid, respectively (78,79). A common mechanism of tumor promotion involves the production of indoleamine 2,3-dioxygenase by DC, which induces tumor tolerance via T-cell suppression. A study by Kim et al. (80) revealed the ability of coumaric acid to decrease the levels of indoleamine 2,3-dioxygenase in DC by blocking the STAT1 signaling pathway, resulting in the release of T cells from suppression. A study by Sierra et al. (81) also revealed the capacity of rice bran oil to modulate the immune response by enhancing B-cell proliferation and inducing IL-2 and TNF- $\alpha$  production. Additionally, the rice bran extract MGN-3 has been shown to enhance DC maturation (40), induce cytokine production, and increase NK-cell activity (38). Unpublished studies from our laboratory have also shown that rice bran inhibits TNF $\alpha$ -induced NF- $\kappa$ B activation in human CRC cells. Moreover, we observed a decrease in the IL-4-induced expression of CD44, which, along with its variant form (CD44 v3-v6), plays an essential role in the adhesion and infiltration of inflammatory cells in CRC.

**Influence of microbial transformation.** An emerging area of cancer chemopreventive research involves the investigation of the intestinal bacterial communities (microbiota) and the interplay with the intestinal mucosa for the prevention of CRC initiation and progression. Several of the bioactive components found in rice bran are prebiotic in nature, including a variety of nondigestible carbohydrates (e.g., pectins, arabinoxylan, lignin, cellulose, and hemicellulose) (82) and soluble fibers (e.g.,  $\beta$ -glucan and gum) (64). These rice bran prebiotics may shape the microbiota toward beneficial bacteria based on the evidence of increased native gut *Lactobacillus* spp in mice (77). Fermentation of these components can affect the intestinal environment through the release of microbial byproducts known as SCFA. SCFA, such as acetate, butyrate, and propionate, have been shown to enhance

the health of colonic enterocytes as well as interact with mucosal immune cells (83–85). Studies in our laboratory revealed that dietary rice bran fermented with *Saccharomyces boulardii* inhibited the growth of Raji B cell lymphoma cells (18). A study by Komiyama et al. (86) revealed the utility of the insoluble portion of rice bran to protect the mucosal epithelium from damage via an increase in the intestinal levels of the microbial metabolites acetate and butyrate. Additionally, the use of pectin suppresses colonic tumorigenesis (87). Pectin-induced chemoprevention may stem from fermentation by a variety of lactic acid bacteria species in vivo, resulting in the increased production of acetic acid (88,89). A study by Snart et al. (90) found that the supplementation of  $\beta$ -glucan in the diet of rats increased the overall proportion of *Lactobacillus* spp within the cecal microbiota. Finally, the hemicellulose portion of rice bran was found to lower the tumor incidence in 1,2-dimethylhydrazine-induced CRC rat study. The authors found that the hemicellulose diet altered the gut microflora populations and decreased the overall intestinal pH with the increased SCFA (91).

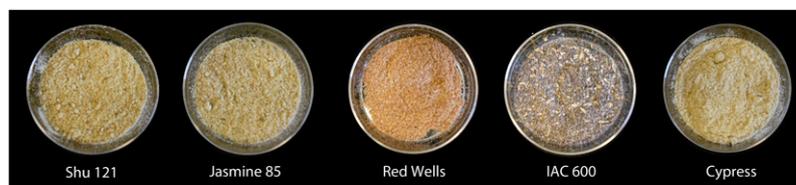
Increasing evidence suggests that populations of cancer stem cells are present in most neoplasms, and early events in cancer initiation involve the expansion of these cancer stem cells. Phytochemicals, such as those present in rice bran, have been shown to induce apoptosis and block differentiation of cancer stem cells (92).

Colonic bacterial species also have the capacity to produce enzymes known for cleaving procarcinogenic compounds into carcinogens. A study by Gestel et al. (93) revealed the ability of rice bran-derived components to protect the colonic epithelium via its ability to decrease the activities of the adverse bacterial enzymes  $\beta$ -glucuronidase and mucinase. Additionally, a study in which rats were fed a diet supplemented with cellulose showed a decrease in the nitroreductase,  $\beta$ -glucuronidase, and  $\beta$ -glucosidase enzyme activity in the colon (94). Therefore, there are a number of mechanisms that are of importance to consider when evaluating the potential efficacy of rice bran for dietary cancer chemoprevention.

### Summary and future prospects

Dietary rice bran and its associated bioactive compounds have been shown to possess immense potential as cancer chemopreventive agents in animal and cell culture studies. In light of the studies reviewed here, the continued investigation into the multitude of rice varieties worldwide and their unique bioactive components holds great promise for optimizing rice bran for future use as a dietary cancer chemopreventive agent (Fig. 3). Further studies on the

**Figure 3.** Visual examples of bran polished from distinct rice varieties that contain unique pigments and bioactive components with cancer chemopreventive potential.



bioavailability, metabolism, and tissue disposition of the rice bran bioactive components in humans are needed.

In conclusion, the continued research into dietary brown rice and rice bran as a cancer chemopreventive food represents a unique opportunity to explore effective whole food approaches compared with single-agent supplements for cancer chemoprevention. Our review of encouraging results from animal and cell culture studies warrants the design of clinical studies to better understand the relationship between whole-grain rice/rice bran and cancer. Moreover, the global availability, accessibility, and affordability of dietary rice bran offer unique public health opportunities in both developed and developing countries.

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