# **Review Article**

# **Cereals:** from staple food to nutraceuticals

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**Abstract:** Natural products such as cereals are likely to form the basis of nutraceuticals. The nutraceutical revolution represents an enormous opportunity for growth and expansion. Wheat, buckwheat, oat, barley, flaxseed, psyllium, brown rice, soy and products are notified the most common cereal based functional foods and nutraceuticals. The nutrients in the cereals have known potential for reducing the risk of coronary heart disease, reducing tumor incidence, cancer risk, lowering blood pressure, reduces the rate of cholesterol and fat absorption, delaying gastrointestinal emptying and providing gastrointestinal health. Thus diet with the regular inclusion of cereals can contribute much to health promotion and disease prevention.

### Keywords: nutraceuticals, cereals, disease, functional food

#### Introduction

The term "nutraceutical" was coined from "nutrition" and "pharmaceutical" in 1989 by Stephen DeFelice, M.D, founder and chairman of the Foundation for Innovation in Medicine (FIM), Cranford, according to him, nutraceutical can be defined as, "a food (or part of a food) that provides medical or health benefits, including the prevention and/or treatment of a disease." However, the term nutraceutical as commonly used in marketing has no regulatory definition (Zeisel, 1999). Nutraceuticals may range from genetically engineered foods to dietary supplements, herbal formulations and may even include processed products like cereals, soups and beverages. Cereals are an important economic commodity worldwide. Food ingredients from cereals with nutraceutical properties can contribute to health benefits to many people. Consumption of plant-based foods, including fruits, vegetables and whole grains, cereals and nuts as well as intake of marine foods plays a pivotal role in disease prevention and health promotion. Cereals like wheat, maize, rice, oats etc are now employed in preparation of foods that are similar in appearance to conventional foods and used in normal diet but have an added advantage of aiding physiological functions along with providing nutrition. Eating habits can drastically reduce healthcare expenditures if individuals were to modify their diets based on an existing knowledge of nutrition.

This review focuses on some commonly consumed cereals with the potentiality of being considered as nutraceuticals.

#### Rice

Rice (Oryza sativa L.) a major cereal crop is the staple food sources for half of the world population. Rice is an important source of energy, hypoallergenic, easily digested, providing protein with higher nutritional quality and has versatile functional properties. The rice kernel comprises 20% hull, 8-12% of bran and embryo and 70-72% endosperm or milled rice based on the degree of milling. Rice bran is a valuable by product of rice milling that contains a high concentration of nutritional compounds, including edible lipids. The rice grain contain 5% bran, of which 12-18.5% is oil. rice bran oil is not popular worldwide, but in demand as known as healthy oil. Rice bran oil contains a range of fats, with 47% of its fats monounsaturated, 33% polyunsaturated, and 20% saturated. A major rice bran fraction contains 12%-13% oil and highly unsaponifiable components (4.3%). This fraction contains tocotrienols (a form of vitamin E), gammaoryzanol, and beta-sitosterol; all these constituents may contribute to the lowering of the plasma levels of the various parameters of the lipid profile. Rice bran also contains a high level of dietary fibers (betaglucan, pectin, and gum). In addition, it also contains 4-hydroxy-3-methoxycinnamic acid (ferulic acid),

Fatty acid	Percentage
Palmitic acid	15.0%
Stearic acid	1.9%
Oleic acid	42.5%
Linoleic acid	39.1%
Linolenic acid	1.1%
Arachidic acid	0.5%
Behenic acid	0.2%
	Palmitic acid Stearic acid Oleic acid Linoleic acid Linolenic acid Arachidic acid

The fatty acid composition of rice bran oil: (Source: "Rice Bran Oil". Retrieved 2006-10-09)

Composition of rice bran: (Source: Wells, 1993)

Serial No	Constituent	Percentage
1.	Protein	11.5-17.2%
2.	Starch*	10-55%
3.	Ash*	8-17.7%
4.	Fiber	6.2-31.5%
	Soluble fiber	1.9-2.4%
	Insoluble fiber	4.4-29%
5.	Phosphorus**	1.5-1.7%
6.	Potassium	1.4-1.5%
7.	Magnesium	0.78%
8.	Tocopherol (brown rice)	22-31 ppm
9.	Calcium	0.02%
10.	Oryzanol (brown rice)	500-720 ppm
11.	Tocotrienol (brown rice)	2-26 ppm

\*- based on the extraction rate

\*\* in the form of phytate

Composition of  $\gamma$  oryzanol (Source: Berger, 2005)

Serial No	Constituents	Percentage
1.	Cycloartenyl ferulate	33%
2.	24-methylenecycloartenyl ferulate	27%
3.	Cyclobranyl ferulate	2%
4.	Campesteryl ferulate	24%
5.	Sitosteryl ferulate	11%
6.	Stigmasteryl ferulate	2%

which is also a component of the structure of nonlignified cell walls.

### Rice bran and rice bran oil as functional food

Rice bran and cardiovascular diseases: Rice bran contains primarily both soluble and insoluble fiber. Insoluble fiber adds bulk to gastrointestinal track in human causing more frequent stools that pass through the system more quickly, requiring less pressure to expel, and absorbing more bile acids and prevents their reabsorption to the body (Wells, 1993). This leads to the synthesis of more bile acids from available cholesterol. Rice bran lowers the serum cholesterol levels in the blood, lowers the level of bad low density lipoprotein (LDL) and increases the level of good high density lipoprotein (HDL) level, aids in cardiovascular health. LDL/HDL ratio is a reliable marker for coronary heart diseases, higher the ratio more will be the risk of coronary heart diseases. RBO significantly lowered the LDL/HDL ratio by 19% over 4 weeks, being equally effective after each 2 week period (Berger et al., 2005).

Rice bran contains phosphorus, potassium, magnesium, calcium, manganese and other trace elements. Magnesium improves glycemic control and helps prevent insulin resistance. Rice bran also contain alpha lipoic acid, which can assist in metabolizing carbohydrates and fats thus called Metabolic antioxidants, which lowers glycemic index and controls body weight. Other than alpha lipoic acid, rice bran also contain energy boosting phytonutrients like CoQ10 and B vitamin including pangamic acid. They are vital for energy metabolism and electron transport in the mitochondria and serves as an effective intercellular antioxidant. CoO10 improves energy in the heart muscles.it provides the body with the energyit needs to rebuild and repair, remove cellular waste and toxins, and itself from infection.Gamma oryzanol is also found to be affective in burning fat, metabolizing stored fat and increasing lean body mass. Studies have shown that 7% oryzanol rice bran oil concentrate to diets of rats shows reduction in bone loss and risk of osteoporosis.

Rice bran contains both Lutein and Zeaxanthin, which improves eyesight and reduces the chance of cataracts. The essential fatty acids, omega-3, omega-6, omega-9 and folic acid in rice bran are also promotes eye health. Vitamin K and Inositol Hexaphosphate (IP6) plays a vital role in preventing kidney stones. Vitamin K moves the stone producing calcium out the blood stream and get into our bones. Inositol Hexaphosphate inhibits urinary calcium oxalate crystallization. Inositol, phospholipid

and Vitamin B complex detoxifies the liver, control liver cirrhosisand improve cell regeneration. Fibers also reduces and removes the toxics and at optimum pH improves digestive function and allows frequent stool pass leads to preventing colon cancer.

Rice bran is potentially a valuable source of natural antioxidants such as tocopherols, tocotrienol and oryzanol (Godber and Wells, 1994). Increased concern over the safety of synthetic antioxidants like butylated hydroanisole (BHA) and butylated hydroxytoluene (BHT) has increased the interest in finding effective and economical natural antioxidants. Antioxidants extracted from rice bran potentially could satisfy this demand. Results from studies have shown that rice antioxidants at 500ppm provide the same level of antioxidant activityas a mixture of BHA/BHT at 200 ppm (Hettiarachchy et al., 993). Reactive oxygen species (ROS) are free radicals produced during metabolism and the ageing process. ROS includes superoxide anion, hydrogen peroxide and hydroxyl radicals. ROS can cause DNA strand breaks, base modification, lipid peroxidation, and protein modification, resulting in oxidative stress. Oryzanol, tocopherol and tocotrienol of rice bran prevents and oxidative stress as well as lipid oxidation (Chiang An-Na, 2006).

## Functional properties of oat and barley, $\beta$ -glucan

Barley is the worldwide used traditional food grain and also historically connected with malt and beer production. Barley is preferred not only for its nutritional importance but also for its nutraceutical properties. The common oat (Avena sativa) is a species of cereal grain grown for its seed, which is known by the same name (usually in the plural, unlike other grains). While oats are suitable for human consumption as oatmeal and rolled oats, one of the most common uses is as livestock feed. The active component in oat and barley having nutraceutical property is the soluble fibre  $(1\rightarrow 3)(1\rightarrow 4)$ -  $\beta$  -Dglucan or  $\beta$ -glucan.  $\beta$ -glucan are polysaccharides found principally in the cell walls of the aleurone layer and endosperm in barley and oat kernels. In barley they are more concentrated in the endosperm while in oats they are concentrated in the aleurone layer (Bhatty R.S, 1993). The largest (seed) amounts of  $\beta$ -glucan are found in barley (3-11%) and oats (3-7%), with lesser amounts reported in rye (1-2%)and wheat (<1%). Only trace amount have been reported in corn, sorghum, rice and other cereals of importance as food (Wood, 1992).

The Food and Drug Administration has made a health claim between  $\beta$ -glucan and reduced risk of coronary heart disease, diabetes and heart related

problems. Studies have shown that the feeding of 140 g of rolled oats to healthy young men lowered serum cholesterol by 11% in three weeks (de Groot et al., 1993). 7.2 g of 80%  $\beta$ -glucan containing oat gum can reduce the LDL and total cholesterol levels by 10% and 9.2% respectively, in 4 weeks (Hohner and Hyldon, 1977). β-Glucan is a soluble fibre and has the ability to increase solution viscosity and a likely rapid fermentation in the small intestine. β-Glucan can delay gastric emptying, increases gastro intestinal transit time and luminal viscosity, and increases thickness of unstirred layer. These characteristics of  $\beta$ -glucan are associated slowed nutritient absorption, reduced blood glucose and insulin level. Short chain fatty acids (acetic acid, propionic acid and butyric acid) produced by fermentation in the colon may also influence the production of glucose and its utilization by peripheral tissues (Jenkins, 1995). Short chain fatty acids may modify cholesterol synthesis (Bridges, 1992). In hypercholesterolemic subjects with ileostomy, wih a virtually suppressed fermentation process, oat bran produced a significant reduction of serum total and LDL cholesterol comparable to that measured in human subjects with a functional colon (Zhang, 1992).

Soluble fibres, in particular, are thought to exert a preventive role against heart diseases as they appear to have the ability to lower serum cholesterol levels. There are several possible mechanisms by which  $\beta$ -glucan is thought to reduce serum cholesterol levels; many are related to the ability of soluble fibres to form viscous gel in the intestinal tract. It has been proposed that  $\beta$ -glucan reduces plasma cholesterol through its ability to bind acids in the gastrointestinal tract. As  $\beta$ -glucan bind bile acid in the intestinal tract, the bile cid level decreases in the body and thus the dietary cholesterol is utilized for the synthesis of bile acids by the liver. Insulin plays a role in lipid metabolism and may stimulate cholesterol synthesis (Bhathena, 1974) and hepatic synthesis and secretion of low density lipoproteins (Reaven G.M., 1978).  $\beta$ -Glucan reduces the rise in the blood glucose and lowers insulin level, which may lead to lower serum cholesterol levels. β-Glucan may bind or inactivate pancreatic lipase and in gastric conditions  $\beta$ -glucan significantly lowers the extent of lipid emulsification and triglyceride hydrolysis catalysed by gastric lipase.

The production of SCFA during the fermentation of  $\beta$ -glucan in the colon is associated with the stimulation of colonic cell proliferation, which has been associated with enhanced tumorigenesis (Zhang, 1994). Regular doses of  $\beta$ -glucan to rats can reduce the risk of 1,2-dimethylhydrazine(DMH) induced cancer (McIntyre, 1993). The ability of  $\beta$ -glucans to bind metabolites, nutrients or carcinogens, or their interference with such binding, clearly could have great physiological significance and play an important role in maintaining the human health.

# Wheat

Wheat is one of the major grains in the diet of vast number of the world's population and, therefore, can play an important role in the nutrition quality of the diet and human health. Like other typical cereal grains, wheat kernel contain three main anatomical parts- an embryo, an endosperm, and pericarp that cover the endosperm. The outermost bran layers are fibre rich. Starch and proteins are concentrated in the endosperm, while the germ is high in fat. The bran and germ fraction are also high in vitamins and minerals.

Wheat bran in particular, widely available as a food ingredient, is probably is the most studied fraction of the grain due to its role is enhancing health and the management and risk reduction of some chronic diseases. Whole wheat and wheat bran are the important source of dietary fibre and antioxidants. Phenolic acids in whole wheat bran having strong antioxidant activity invitro at concentration that obtained from a normal serving of whole wheat cereal. Acid conditions and enzymatic hydrolysis increases the solubility and activity of wheat phenols (Baublis et al., 2000). Whole grains are a rich source of magnesium, a mineral that acts as a co-factor for more than 300 enzymes, including enzymes involved in the body's use of glucose and insulin secretion. The FDA permits foods that contain at least 51% whole grains by weight (and are also low in fat, saturated fat, and cholesterol) to display a health claim stating consumption is linked to lower risk of heart disease and certain cancers. Now, research suggests regular consumption of whole grains also reduces risk of type 2 diabetes. (van Dam and Hu, 2006).

The useful role of wheat bran in promoting regulating and preventing constipation is generally accepted. In addition, growing research has focused on its protective effect against colon and breast cancers. Constipation is the infrequent bowel action, twice a week or less. Low fibre content in diet is the major factor in the development of constipation. In some instances, complete relief of constipation may not be achieved with dietary fibre supplementation alone. According to Health and Welfare Canada, constipation can be prevented with supplemental fibre intake from wheat bran has been shown to be an effective strategy in up to 60% of elderly patients and in children. Wheat bran is a popular bulk laxative. A third of a cup per day is all that is needed. Research studies support this popular practice. A fiber-rich diet, primarily composed of whole wheat breads, cereals high in bran and supplemental "millers bran" was shown to alleviate the symptoms of diverticular disease (pain, nausea, flatulence, distension, constipation, etc.) in 89 percent of patients.

Wheat bran is thought to accelerate the metabolism of estrogen that is a known promoter of breast cancer. Pre-menopausal women, ages twenty to fifty, who ate three to four high fiber muffins per day made with wheat bran, decreased their blood estrogen levels by 17 percent after two months. The women eating corn bran or oat bran did not show the same benefits (Suzuki et al., 2008). Wheat also contain lignans, which are phytonutrients that act as weak hormonelike substances. Lignans occupy the hormone receptors in the body, thus actively protecting the breast against high circulating levels of hormones such as estrogen. The benefit of diet characterized by high fibre food in colon cancer risk reduction has been well recognised although the dietary component involved has not been clearly defined. There is a significant inverse relationship between measures of fibre intake and colon cancer risk, but all fibres are not equally involved in n their protective action against colon cancer. Only the bran from wheat has been shown to reduce the concentration of bile acids and bacterial enzymes in the stool that are believed to promote colon cancer. Among the all fibre sources, wheat bran appears to have the most consistent inhibiting effect on colon cancer development.

# Millet

Millet is one of the oldest foods known to humans and possibly the first cereal grain used for domestic purposes. The millets are a group of small-seeded species of cereal crops or grains, widely grown around the world for food and fodder. The protein content in millet is very close to that of wheat; both provide about 11% protein by weight. Millets are rich in B vitamins, especially niacin, B6 and folic acid, calcium, iron, potassium, magnesium, and zinc. Millets contain no gluten, so they are not suitable for raised bread. When combined with wheat, (or xanthan gum for those who have coeliac disease), they can be used for raised bread. Millet is highly nutritious, non-glutinous least allergenic and most digestible grains. The seeds are also rich in phytochemicals, including Phytic acid, believed to lower cholesterol, and Phytate, which is associated with reduced cancer. However, millets are also a mild thyroid peroxidase inhibitor and probably should not be consumed in great quantities by those with thyroid disease (Crawford and Gary, 2003).

Corn, also called maize, is a cereal grass related to wheat, rice, oats, and barley. Corn is known scientifically as Zea mays. corn as a good source of many nutrients including thiamin (vitamin B1), pantothenic acid (vitamin B5), folate, dietary fiber, vitamin C, phosphorus and manganese. Corn's contribution to heart health lies not just in its fiber, but in the significant amounts of folate that corn supplies. Corn maintains the homocysteine, an intermediate product in an important metabolic process called the methylation cycle. Homocysteine is directly responsible for demage of blood vessel heart attack, stroke, or peripheral vascular disease. It has been estimated that consumption of 100% of the daily value (DV) of folate would, by itself, reduce the number of heart attacks suffered by 10%. (Bazzano et al., 2002). Corn also contains cryptoxanthin, a natural carotenoid pigment. It has been found that cryptoxanthin can reduce the risk of lungs cancer by 27% on daily consumption (Yuan, 2003). Adom and Liu (2002) analyzed a number of cereals, namely corn, wheat, oat and rice and reported that corn had the highest free phenolic content (0.411 mg/g of grain), followed by rice (0.407 mg/g of grain), then wheat (0.368 mg/g of grain), and oat (0.343 mg/g of grain). The content of insoluble-bound phenolic was significantly higher among all of the above cereals. Phenolic compounds of corn may render their effects via antioxidation and relief from oxidative stress and its consequences. The antioxidative effect of phenolics in functional foods is due to a direct free radical scavenging activity (Halliwell, 1996; Shahidi, 2000; Shahidi and Ho, 2007), reducing activity and an indirect effect arising from chelation of prooxidant metal ions.

## Sorghum

Sorghum is an annual grass that is extremely drought tolerant, making it an excellent choice for arid and dry areas. Sorghum has special adaptations to weather extremes and is a very stable source of nutrition as a result. Sorghum is favored by the gluten intolerant and is often cooked as porridge to be eaten alongside other foods. The grain is fairly neutral in flavor, and sometimes slightly sweet. Sorghum is commonly eaten with the hull, which retains the majority of the nutrients. The plant is very high in fiber and iron, with a fairly high protein level as well.

3-Deoxyanthocyanidins are structurally related to the anthocyanin pigments, which are popular as health-promoting phytochemicals. Sorghum is the only dietary source for 3-deoxyanthocyanidins, which are present in large quantities in the bran of some cultivars (Awika, 2004). The defense mechanism of sorghum against pathogen is due to an active process, resulting in the accumulation of high levels of 3-deoxyanthocyanidin phytoalexins in infected tissues (Lo et al., 1996).

Luteolinidin and apigeninidin are the two major 3-deoxyanthocyanidins, and they are structurally related to anthocyanidins. Both luteolinidin and apigeninidin exhibited stronger cytotoxicity on the (Human leukemia) HL-60 cell. Luteolinidin reduced the viability of HL-60 cells by >90% at 200  $\mu$ M, whereas cyanidin reduced the viability by only around 20% at the same concentration. Apigeninidin was less effective than luteolinidin (around 70% reduction at 200  $\mu$ M). Overall, cytotoxic activities of the 3-deoxyanthocyanidins on the hepatoma HepG2 cells were not as effective as they were on the HL-60 cells. At 200  $\mu$ M, luteolinidin and apigeninidin reduced the viability of the cancer cells by around 40% (Shih Chun-Hat et al., 2007).

## Buckwheat

Buckwheat (Fagopyrum esculentum Moench, Polygonaceae) is a crop which holds tremendous agronomic and nutritional benefits. Buckwheat is one of the best sources of high quality, easily digestible protein in the plant kingdom. It has over 90% of the value of non-fat milk solids and over 80% of whole egg solids (Udesky, 1992). The balanced amino acid profile and a high level of essential amino acids allow buckwheat to be used in human diets, especially where shortages of lysine and sulfur containing amino acids appear. . The main protein solubility fraction in buckwheat is globulin. In addition, the content of prolamines is low. It is relatively high in potassium and phosphorus, and contains 150% more vitamin B than wheat. Buckwheat has no more calories than wheat products or most other grains. It is gluten free, thus making it a valuable nutrient in the diets of people who are sensitive to gluten. Buckwheat contains a glucoside named rutin, a medicinal chemical that strengthens capillary walls, reducing hemorrhaging in people with high blood pressure and increasing microcirculation in people with chronic venous insufficiency (Ihme N., 1996). Buckwheat contains vitamin P, which contains the flavonoid rutin. Rutin is known for its effectiveness in reducing the cholesterol count in the blood. In addition, buckwheat is an effective preventative measure against high blood pressure. Rutin is known to keep capillaries and arteries strong and flexible. The effectiveness of rutin in buckwheat is strengthened with the addition of vitamin C (Udesky, 1992). Buckwheat contains considerable amounts of vitamins B1 and B2. Vitamin B1 is important in rekindling energy by facilitating the working of the nerves. Vitamin B2 assists the lipids in their work. Combining these vitamins with vitamin E, results in an effective precaution against hardening of the arteries (arteriosclerosis). Potassium, magnesium, phosphate, and iron are abundant in buckwheat flour. Buckwheat is higher in iron than cereal grains. These minerals play an essential role in the prevention of hypertension and anemia (Udesky, 1992).

# Processed cereals and their nutraceutical properties

Starch is the major storage carbohydrate of all higher plants and is the major constituent of cereals. The physical, chemical and enzymatic processing of cereals modifies the characteristics of starch, which impart the nutraceutical properties. However, it has been found that a part of starch consumed in the diet escape digestion and absorption in the small intestine and is fermented in the large intestine with the production of short-chain fatty acids (Asp, 1992). This fraction is named resistant starch (RS). RS has been associated with a reduction of the glycemic index, low absorption of cholesterol, and prevention of colon cancer (Englyst, 1992). RS is found naturally in a broad range of starchy products and can be added as a functional ingredient. Resistance starch concentration of cereals increases during processing. Process heating and cooling cycles are used for promoting starch retrogradation which, in turn, increases RS content. Another method for RS production involves starch gelatinization, enzymatic debranching of the gelatinized polymer, deactivation of the debranching enzyme, and isolation of the resultant product either by drying, extrusion, or crystallization. Extrusion has been claimed as a unit operation for RS production; however, the RS levels obtained with this procedure are lower than those prepared using autoclaving (Agustiniano-Osornio, 2005). The nutraceutical potential of starchy products with low or reduced starch digestion is related with a reduction in the availability of starch to digestive enzymes (a-amylase, isoamylase, pullulanase), which decreases postprandial blood glucose and insulin response. Other factors may be associated with the decrease of the metabolic response such as a reduced rate of gastric emptying, a reduced motility of the luminal content, and a reduced rate of diffusion of starch hydrolysis products to the small intestinal mucosa (Bjorck et al., 1994). The nutraceutical characteristics associated with the fermentation of the undigestible starch (RS) and the physiological

effects are briefly remarked in the next section; however, there is another nutraceutical property associated to the fermentation, which is the prebiotic effect. RS may also act as a prebiotic because it favorably influences the ecology of the microbial flora in the large intestine (Thompson, 2007). RS can be beneficial with respect to probiotics since the ingestion of RS serves to extend the viability of some probiotic organisms that exist in the colon. When RS and probiotics are consumed together, a symbiotic effect takes place; a prebiotic role of RS is to protect some of the ingested organisms on their hazardous path to the colon, effectively increasing the initial levels of the desirable species, once the colon is reached. When both the probiotics and the RS are present in the colon, then the RS may initiate its role as susbtrate for a portion of the probiotic organisms (Topping, 2003).

Monascus-fermented rice has traditionally been used as a natural food colorant and food preservative of meat and fish for centuries. The genus Monascus is considered to belong to the family Monascaceae, the order Eurotiales, the class Ascomycetes, the phylum Ascomycota, and the kingdom Fungi. It has recently become a popular dietary supplement because of many of its bioactive constituents being discovered, including a series of active drug compounds, monacolins, indicated as the 3- hydroxy-3-methylglutaryl-coenzyme A reductase inhibitors for reducing serum cholesterol level. Monascusfermented rice is described as a mild folk medicine, which has the therapeutic effect to promote the health of cardiovascular system. Among the bioactive compounds found in MFR, monacolins are well known for their pharmacological effects to control hyperlipidemia. Among the monacolins, monacolin K is considered the most efficacious compound to lower cholesterol in the plasma (Lin Yii-Lih., 2008). Dihydromonascolins is an another active compound derived from the methanolic extract of M. purpureus. It contained strong 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging activity and inhibition of lipid peroxidation in a liposome model (Dhale et al. 2007). Dimerumic acid is an antioxidant present in Monascus-fermented rice was further found to inhibit the NADPH- and iron(II)-dependent lipid peroxidation of rat liver microsomes at 20 and 200 μM, respectively (Taira et al. 2002).

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