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Percent Reduction Utilizing Ferric Reducing Antioxidant Power and Levels of Stress from Red and Black Rice

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ABSTRACT

Antioxidants are anti-inflammatory which could be used as therapy for certain inflammatory-linked diseases such as autoimmune diseases, asthma, hepatitis, glomerulonephritis and coeliac diseases. Heart disease, stroke and aging are also leading causes of death which can be reversed through antioxidant intake. Several types of antioxidants have differing properties and knowing how each type affects health in specific doses is essential in combating diseases and maintaining a healthy state. Six groups of antioxidants will be reviewed in this experiment: phenolic acids, flavonoids, anthocyanins, proanthocyanidins, tocopherols, tocotrienols, y-oryzanol and phytic acid. In this experiment, iron reduction will be tracked and analysed using ferric reducing antioxidant power assay. In 700 nm, spectrofluorimetry will be utilized to detect absorbance in this spectrum. Data obtained will be calculated using formula for percent reduction. Antioxidant activity will be determined as there is a direct correlation between reduction and antioxidant activity. Researchers found that percent of reduction was greatest in red rice in 1000 ug/mL with a value of 56.181% compared to 500 ug/mL with 30.53%, 250 ug/mL with -1.35% and 125 ug/mL with 4.30%. In black rice, percent reduction was highest with 1000 ug/mL with a value of 66.20%, 500 ug/mL with a value of 55.87%, 250 ug/mL at 38.51% and 125 ug/mL at -15.07%. Thus, black rice has more reduction compared to red rice, according to data.

Keywords: Antioxidants, Red rice, Black rice, Stress, Reduction

INTRODUCTION

The nutrition of rice is the focus of this research experiment. Rice has about 345 Cal/100 g. It contains vitamin B1, niacin, vitamin D, calcium, fibre, iron, thiamine and riboflavin. Most importantly, the antioxidants present in rice will be the concentration of this research as it could be used as therapy to leading causes of death such as heart disease, stroke, respiratory diseases and cancer. Antioxidants are compounds that inhibit oxidation. Oxidation is a chemical reaction that transfers electron or hydrogen from substances to an oxidizing agent. Oxidation reactions produce free radicals — unpaired electrons which start a chain reaction that causes cell death. Antioxidants found in fruits and vegetables terminate these chain reactions by removing free radical intermediates and inhibit other oxidative reactions [1].

Antioxidants have 2 modes of action: hydrogen atom transfer measure the capability of antioxidant to quench free radicals peroxyl radicals considered to be biologically more relevant by H-atom donation. In SET-based assays, antioxidant action is simulated with a suitable redox-potential probe, antioxidants react with a fluorescent or colored probe (oxidising agent) instead of peroxyl radicals [1].

These compounds, antioxidants, reverse aging, prevent cancer, heart disease, stroke, and Alzheimer's disease which are leading causes of death in the world. Antioxidants reduce inflammation thereby also preventing other inflammatory diseases such as allergy, asthma, autoimmune diseases, coeliac disease, glomerulonephritis, hepatitis and inflammatory bowel disease.

Black and red rice both contain antioxidants in differing amounts. Antioxidants compounds in rice were classified into six groups: phenolic acids, flavonoids, anthocyanins, proanthocyanidins, tocopherols and tocotrienols (vitamin E), y-oryzanol and phytic acid [2]. Phenolic acids contain a

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phenolic ring and an organic carboxylic acid function with absorption maxima at 280 nm for the C6-C1 skeleton of hydroxybenzoic acid derivatives (gallic, protocatechuic, p-hydroxybenzoic, vanillic and syringic acids) and at 320 nm for the C6-C3 skeleton of hydroxycinnamic acid derivatives (p-coumaric, ferrulic, caffeic, sinapic, chlorogenic and cinnamic acids). The phenolic ring can stabilize and delocalize unpaired electrons, conferring an antioxidant property to phenolic acids. Antioxidant property notably depends on the number and the position of hydroxyl groups on the phenolic ring [3]. Phenolic acids are synthesized from the shikimate pathway from L-phenylalanine or L-tyrosine. Phenylalanin and tyrosine are very important amino acids in this pathway since these amino acids are the common precursors of the majority of natural phenolic products [4]. Flavonoids are synthesized by the phenylpropanoid metabolic pathway; most flavonoids have absorption maxima at 370 nm. Flavonoids consist of a 15-carbon skeleton that is organized in two aromatic rings (A and B-rings) interlinked by a three-carbon chain (structure C6-C3-C6). Flavonoids are recognized for both their ability to donate electrons and to stop chain reactions. These activities are attributed to phenolic hydroxyls, particularly in the 3'OH and 4'OH of the three-carbon chain. Flavonoids can be classified into flavones, flavonols, flavanols, flavanonols, isoflavones and flavanones which generally occur as O- or C-glycosides. Of the seven flavonoids that are usually reported in rice, tricin appears to be the major flavonoid in the bran, accounting for 77% of all seven flavonoids. Anthocyanins, another class of antioxidants exhibit maximum absorbance in the green/blue spectrum at 510 nm, are water-soluble glycosides of polyhydroxyl and polymethoxyl derivatives of 2-phenylbenzopyrylium or flavylium (2-phenylchromenylium) salts. They share a common hydroxylation at the C3, C5 and C7 positions on the B-ring. Anthocyanins exist as O-glycosides (mono, di, or tri) and acylglycosides of anthocyanidins in plants. The sugars may be substituted by aliphatic, hydroxybenzoic or hydroxycinnamic acids. The structural characteristics of anthocyanins make them highly reactive toward reactive oxygen species. Tocotrienols and tocopherols share a common structure unit based on amphiphilic 6-chromanol ring and a terpenoid side chain located at position 2 of the ring. The chromanol head group can be joined to a saturated phytyl side chain to form tocopherols or to an unsaturated geranylgeranyl side chain to form tocotrienols. The head group can then be methylated in different configurations, resulting in four alternative forms (α, β, γ and ş). The free hydroxyl group on the chromanol ring is responsible for the antioxidant properties and the hydrogen atom from this group can be donated to free radicals, resulting in a resonance-stabilized vitamin E radical [5]. Gamma-oryzanol is a mixture of steryl ferulate (y-oryzanol) which are formed by esterification of the hydroxyl group of sterols (campersterol, stigmasterol, b-sitosterol) or triterpene alcohols (cycloartanol, cycloartenol, 24-methylenecycloartanol, cyclobranol) with the carboxylic acid group of ferulic acid. Sterols with saturated steroid skeleton are known as stanols, whereas compounds containing a double bond between C5 and C6 or between C7 and C8 are referred to as sterols. Methyl groups at C4 affect the antioxidant properties of steryl ferulates [2].

SCIENTIFIC LITERATURE REVIEW

The history of rice dated back in China. According to archeological evidence, rice was domesticated in the region of the Yangtze River valley in China. Rice is the most important food crop and dietary staple, ahead of wheat, corn and bananas. It is the chief source of food for about 3 billion people, half of the world's population, and accounts for 20% of all the calories that mankind consumes. In Asia, more than 2 billion people rely on rice for 60-70% of their calories. If consumption trends continue 4.6 billion people will consume rice in 2025 and production must increase 20% to keep up with demand [6].

In Japan, Korea and other countries, farmers now use small diesel-powered rototiller-tractors to plow the rice paddies and refrigerator-size mechanical rice transplanters to plant the rice seedlings. In the old days it took 25 to 30 people to transplant seedlings of one rice paddy [6].

Water depth in the paddy is increased as rice seedlings grow and gradually lowered in increments until field is dry when rice is ready to be harvested. Water is also drained during growing season so the field can be weeded and soil aerated and water is put back in. Rice is harvested when it is golden-yellow color several weeks after water completely drained from the paddy and soil around rice is dry [6].

Phenolic acids undergo absorption in the gastrointestinal tract then these molecules suffer conjugation reactions and several changes in their initial structure and circulate in human plasma as conjugated forma, glucoronide, methylated and sulphated derivatives. These changes in their structures may increase or decrease bioactivity of the initial phenolic acids [4]. Plant antioxidant activity is due to phenolic acid content, especially caffeic and p-coumaric. The greater antioxidant activity of caffeic acid than p-coumaric acid stems from the 3,4-position of dihydroxylation of the phenolic ring. Phenolics behave as antioxidants due to the reactivity of the phenol moiety (hydroxyl substituent) on the aromatic ring. Caffeic acid is expected to have higher antioxidant activity due to additional conjugation in the propanoic side chain, which facilitates electron delocalization, by resonance, between aromatic ring and proanoic group [7]. Detection of phenolic acids at 280nm is the best alternative for the determination of both classes of phenolic compounds. Extensive use of photodiode array detection in the analysis of phenolic acids can be attributed to the ability to collect online spectra without using stopped-flow techniques [7].

Alpha tocopherol in vitamin E is absorbed via the lymphatic pathway and transported in association with chylomicrons. In plasma alpha-tocopherol is bound to high density lipoproteins. After intestinal absorption and transport with chylomicrons alpha-tocopherol is mostly transferred to parenchymal cells of the liver where most of the fat-soluble vitamin is stored [8]. Alpha-tocopherol inhibits smooth muscle cell proliferation, decreases protein kinase C activity, increases phosphoprotein phosphatase 2A activity and controls expression the alpha-tropomyosin gene. These functions are not related to vitamin E antioxidant action because beta-tocopherol, which has a similar antioxidant activity, does not perform any of these functions; it actually abrogates the alpha-tocopherol effect. Alpha-tocopherol effects on protein kinase C inhibition have been reported in human platelets, diabetic rat kidney and human monocytes. The mechanism of protein kinase C inhibition by alpha tocopherol may be attributable in part to its attenuation of the generation of membrane-derived diacylglycerol, a lipid that activates protein kinase C translocation and activity. Inhibition of protein kinase C activity is not due directly to the antioxidant capacity of alpha-tocopherol, but requires integration of alpha-tocopherol into a membrane structure, likely due to direct interaction between alpha-tocopherol and protein kinase C in cell membrane [9].

Chromanol head group can be joined to a saturated phytyl side chain to form tocopherols, or to unsaturated geranyl-geranyl side chain to form tocotrienols in vitamin E. Free hydroxyl on chromanol ring is responsible for antioxidant properties and hydrogen atom can be donated to free radicals resulting in resonance-stabilized vitamin E radical. Unsaturated tocotrienols found to have greater antioxidant properties and fewer methyl groups lower steric hindrance impending penetration into membranes. Antioxidant activities of vitamin E tocotrienols are mediated through induction of antioxidant enzymes such as superoxide dismutase, NADPH:Quinone oxidoreductase and glutathione peroxidase which quench free radicals such as superoxide radicals. Anti-proliferative activity of tocotrienols are mediated through modulation of growth factors such as vascular endothelial growth factor, fibroblast growth factor, and transforming growth factor beta [10,11].

Y-oryzanol is mainly metabolized in the liver, but it is well known that drugs undergo various kinds of metabolism such as ester-hydrolysis and glucuronidation in the small intestine during absorption. Thus, the intestinal metabolism of y-oryzanol was investigated by analyzing mesenteric venous blood in an in situ intestinal absorption experiment. In the present investigation, it was found that the ester linkage of y-oryzanol was partly hydrolyzed in the intestine during absorption, and the extent of hydrolysis was less than 20% of the radioactivity transferred into the mesenteric vein [12]. The Orz components may be useful to prevent the installation of inflammatory process in allergic reaction, since the non-polar structure of cycloartenyl ferulate proved to be capable of sequestering immunoglobulin E and inhibit the allergic reaction mediated by mast cell degranulation. Several health benefits attributed to Orz due to its anti-inflammatory and antioxidant activities. Presence of inflammation increases ROS production inside the cell, either through NADPH oxidase or the mitochondrial electron transport chain. These reactive molecules are directly related to progression of inflammatory processes, as they induce cell injury and or lead to activation of redox-sensitive transcription factors. Some ROS arising from plasma or organelles membrane can influence transcription by regulating phosphorylation of transcription factors, whereas ROS arising from the perinuclear mitochondria or from a nuclear flavoenzyme can participate in transcriptional control by directly targeting DNA [13].

Flavonoids enter the gastrointestinal tract, afterwards the process of absorption in the small intestine takes place. The degree of absorption depends on several factors and differs among the individual flavonoid subclasses. Highest bioavailability determined for isoflavones, followed by flavanols, flavanones and flavonol glycosides. Flavonoid glycosides are first deglycosylated prior to intestinal uptake, whereas aglycones can freely penetrate through cell membranes. Absorbed flavonoids are transported to the liver where they undergo extensive metabolism generating different conjugation forms such as glucuronides, sulphates and methylated derivatives. Most abundant metabolic reactions of flavonoids are oxidation, reduction, hydrolysis and conjugation with sulphate, glucuronate or O-methylation. These reactions significantly affect the antioxidant activity of flavonoids and their interactions with proteins [14]. Flavonoids inhibit activity of transporters. After metabolites absorbed from small intestine to portal blood, they rapidly reach liver where they undergo several phase 1 and 2 metabolic conversions and in some cases, enter enterohepatic circulation by way of bile and returned to small intestine. Conjugation reactions of flavonoids are catalysed by enzymes UDP-glucuronosyl transferase or sulfotransferase whereas oxidation processes bound to cytochrome P450 enzymes in microsomes [15].

Anthocyanins are transported via the portal vein into the liver and distributed to hepatocytes. After metabolism in the liver, anthocyanins may return to the enteric system through bile or enter general circulation before removal by the kidneys and excretion in urine [16]. Anthocyanins are recognized as dietary polyphenols that are also pigments that color red, blue and purple in fruits and vegetables. Anthocyanins are water-soluble glycosides of polyhydroxyl and polymethoxyl derivatives of 2-phenylbenzopyrylium or flavylium salts. Six anthocyanins commonly found in plants are classified according to the number and position of hydroxyl groups on the flavan nucleus, and are named cyanidin, delphinidin, malvidin, peonidin, pelargonidin and petunidin. Differences between individual anthocyanins come from number and position of hydroxyl groups, degree

of methylation of hydroxyl groups, the nature, number and location of sugars attached to the molecule and aliphatic or aromatic acids attached to the sugars in the molecule. Glycosylation caused increased structural stability and water solubility to the parent anthocyanin. Acylation of sugar residues with cinnamic, caffeic or aliphatic acids further improves stability [17]. Anthocyanin intake ranges between 180 mg/day and their presence in circulation is limited to a few hours [18].

Phytic acid when synthesized is metabolically expensive. It requires six separate ATP-requiring phosphorylations per mole of product. Others have suggested that this substance may serve as a store of phosphorus, of cations, of glucuronate or of high energy phosphoryl groups, which can be metabolized by phytase and phytate-nucleotide diphosphate phosphotransferases during germination to support early events in plant development. Phytate, by virtue of chelating free iron, is a potent inhibitor of iron-driven hydroxyl radical formation. Hydroxyl radical generation mediated by iron requires availability of at least one coordination site that is open or occupied by a readily dissociable ligand such as water. Coordination chemistry of iron phytate chelates with iron-to-phytate ratios exceeding one is unknown due to the low solubility of these polyferric phytate chelates. This completely blocks OH formation and strongly suppresses lipid peroxidation [19].

The transition metal ion Fe^{2+} possesses the ability to perpetuate the formation of free radicals by gain or loss of electrons. Therefore, the reduction formation of reactive oxygen species can be achieved by the chelation of metal ions with chelating agents. This is one of the reasons why Ferric reducing antioxidant power is used in this experiment.

Another mechanism of anti-oxidative action is chelation of transition metals, thus preventing catalysis of hydroperoxide decomposition and Fentontype reactions. In the presence of chelating agents, the complex formation is disrupted with the result that the red colour of the complex is decreased. Measurement of color reduction, allows estimation of chelating activity of coexisting chelator. The main strategy to avoid ROS generation is associated with redox active metal catalysis involving chelating of metal ions [20] **(Figure 1)**.

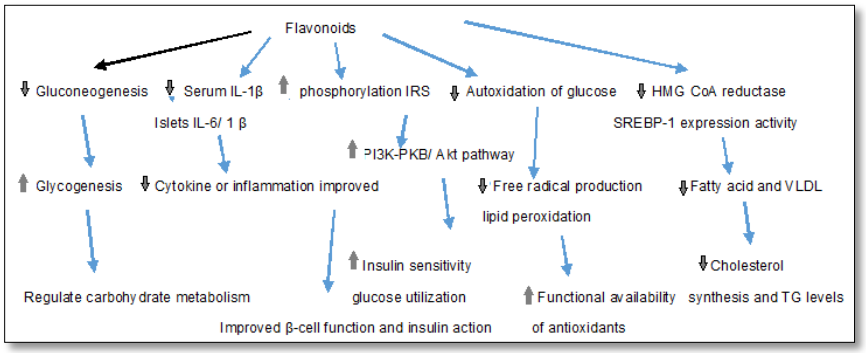


Figure 1. Schematic of role of flavonoids in metabolic reactions [29].

MATERIALS AND METHODS

Extraction

Materials were gathered: 4 test tubes, 1 bottle methanol, 1 mortar and pestle, 1 scooper, 4 Erlenmeyer flasks, 4 1/2 kg bags of red rice and black rice measured at 50 g each and 1 graduated cylinder. 50 g of rice underwent cominution using mortar and pestle. Analytical balance was then used to weigh each sample — red and black rice. Each sample was placed inside Erlenmeyer flask. 120 ml of methanol was measured in graduated cylinder. Sample was slightly shaken — rice and methanol and soaked overnight.

Methanol extracts undergone percolation using filter paper and funnel while filtrate was placed in an Erlenmeyer flask. The filtrate was measured at half of the drum vial to be placed in a rotary evaporator and then replenished until mixture is finished. The sample was processed in 37-40C water bath of rotary evaporator with chiller at 4C until liquid was dry. Each dried extract was measured in an analytical balance by weighing.

Amount of reagents prepared

Sodium phosphate buffer Na_2HPO_4=0.85475192 g, NaH_2PO_4=0.47726086 g

1% Potassium Ferricyanide=0.5 g

10% Trichloroacetic acid=5 g

0.1% Ferric chloride ($FeCl_3$)=0.015 g

50 ml of distilled water was added to each to create a solution except 0.1% ferric chloride (0.015 g) + 15 ml water. pH meter was used for 0.8547 g Na_2HPO_4 + 0.477 g NaH_2PO_4 + 20 ml water.

Ferric reducing antioxidant power assay (FRAP)

FRAP involves the following steps: a) Preparation of samples; b) Reactions; c) Measuring absorbance of sample and standard at 700 nm using spectrofluorimetry. It is the

reduction of ferric to ferrous through an electron transfer process in the presence of antioxidant. High reduction capacity of plant extract would signify that it has an antioxidant activity. 1 mg/mL of dried plant sample of each types of rice — red and black was measured in analytical balance, adding 1000 µl of methanol per sample. This was sonicated for 15 mins. After which, methanol extracts were transferred into 12 microcentrifuge. Each concentration were subdivided into 4 groups — 1000 µg/ml, 500 µg/ml, 250 µg/ml, 125 µg/ml. Per concentration, sample were triplicated. For 1000 µg/ml, concentration was pure. For 500 µg/ml, volume of stock solution would be 250 µl and volume of methanol to be added would be 250 µl, for 250 µg/ml, volume of stock solution was 125 µl and volume of methanol added was 375 µl. For 125 µg/ml, volume of stock solution was 62.5 µl and volume of methanol added was 437.5 µl. This measurement was obtained by using the formula:

$$C1V1=C2V2$$

Seventy microliter (70 uL) of standard (butylated hydroxytoluene) and test compounds at different concentrations were mixed with 176.5 uL of 0.2 M sodium phosphate buffer (pH=7.4) and 176.5 uL of 1% potassium ferricyanide. The mixture was incubated at 50°C for 20 min. After incubation, reaction mixtures were acidified with 176.5 uL of trichloroacetic acid (10%). An aliquot of 136.5 uL of the supernatant was added to 136.5 uL of deionized water. Finally, 27.5 uL of $FeCl_3$ (0.1%) was added to this solution. The absorbance was measured at 700 nm using spectrofluorimeter.

After analysing in the spectrofluorimeter, data was obtained which include absorbance of sample at 700 nm. Percent of reduction was calculated through the formula:

$$(C1-A1/C1) \times 100$$

Each equation was done in triplicate. Thereafter, results were averaged. Data was bar graphed with concentration in µg/mL in x-axis while percent reduction in y-axis.

RESULTS AND DISCUSSION

The table provided the data comparing black and red rice and BHT (butylated hydroxytoluene) which is a synthetic actioxidant to be able to have a standard to compare the levels of antioxidant activity in different amounts of the extracts from black and red rice — 125, 250, 500 and 1000 ug/mL. Each amount was utilized in triplicate thereafter obtaining the average of the three samples. The mean for each sample was then compared and evaluated.

The **Figures 2 and 3** show the shades of green according to antioxidant activity. It is observed that there was greater antioxidant activity upon higher levels of the extract (1000 ug/mL) in both red and black rice. On row A of both **Figures 2 and 3**, the blank sample was colored yellow which indicated no antioxidant activity. The BHT sample from the next rows showed a parallel result that the shade of green was solid since it is a synthetic antioxidant. The extracts from the next column from **Figure 2** showed a darker green comparing to the next rows below it. In **Figure 3**, there is a parallel relationship with **Figure 2**.

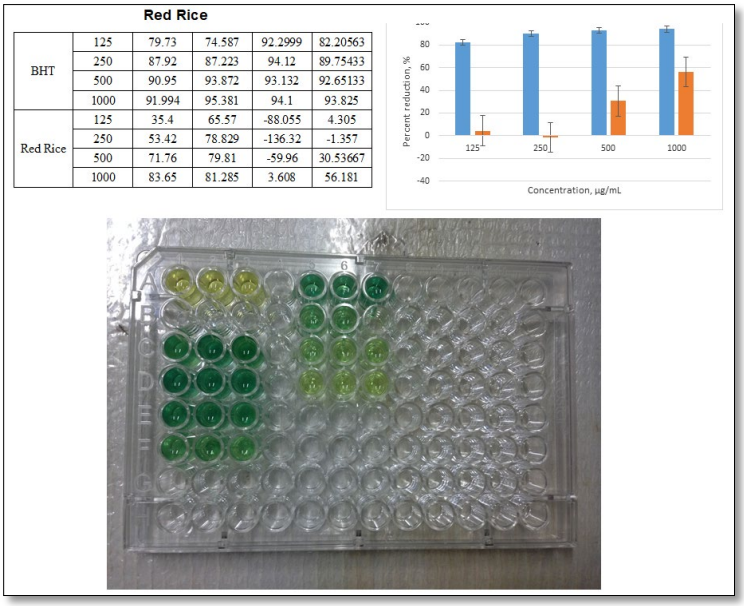
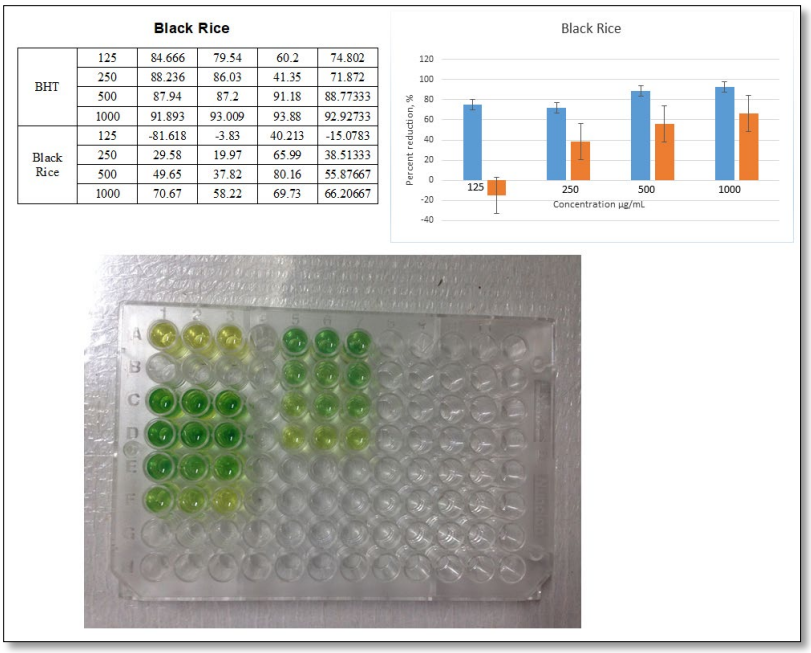


Figure 2. Red rice assay. Darker shade of green shows higher antioxidant activity. A1, A2, A3 — Blank sample. C1, C2, C3 — BHT 1000 ul. D1, D2, D3 — BHT 500 ul. E1, E2, E3 — BHT 250 ul. F1, F2, F3 — BHT 125 ul. A5, A6, A7 — Red rice methanolic extract 1000 ul. B5, B6 — Red rice methanolic extract 500 ul. C5, C6, C7 — Red rice methanolic extract 250 ul. D5, D6, D7 — Red rice methanolic extract 125 ul.



Black Rice

BHT	125	84.666	79.54	60.2	74.802
	250	88.236	86.03	41.35	71.872
	500	87.94	87.2	91.18	88.77333
	1000	91.893	93.009	93.88	92.92733
Black Rice	125	-81.618	-3.83	40.213	-15.0783
	250	29.58	19.97	65.99	38.51333
	500	49.65	37.82	80.16	55.87667
	1000	70.67	58.22	69.73	66.20667

Figure 3. Black rice assay. Darker shade of green shows higher antioxidant activity. A1, A2, A3 — Blank sample. C1, C2, C3 — BHT 1000 ul. D1, D2, D3 — BHT 500 ul. E1, E2, E3 — BHT 250 ul. F1, F2, F3 — BHT 125 ul. A5, A6, A7 — Black rice methanolic extract 1000 ul. B5, B6, B7 — Black rice methanolic extract 500 ul. C5, C6, C7 — Black rice methanolic extract 250 ul. D5, D6, D7 — Black rice methanolic extract 125 ul.

CONCLUSION

In this research experiment, black rice was found to have more reduction compared to red rice. This signifies that black rice has more antioxidant activity compared to red rice. Percent reduction of black rice was 66.2% for 100 ug/mL and 56.181% for 100 ug/mL.

OPERATIONAL DEFINITION OF TERMS

Antioxidants: Antioxidants is a molecule that inhibits the oxidation of other molecules. Oxidation is a chemical reaction that can produce free radicals, leading to chain reaction that may damage cells.

Anthocyanins: Anthocyanins are water soluble vacuolar pigments that, depending on their pH, may appear red, purple or blue. They belong to a parent class of molecules called flavonoids synthesized via the phenypropanoid pathway [21].

Flavonoid: Flavonoid is a group of natural substances with variable phenolic structures found in fruits, vegetables, grains, bark, roots, stems flowers, tea and wine [22].

Phenolic acids: Phenolic acids are defined chemically as carboxylic acids derived from either benzoic or cinnamic acid skeletons [23].

Proanthocyanidins: Proanthocyanidins are oligomeric flavonoids. They are dimers or oligomers of catechin and epicatechin and their gallic acid esters (phy).

Tocopherols: Tocopherols are a group of lipid-soluble constituents composed of a polar moiety derived from tyrosine, the chromanol ring and a hydrophobic phytyl-derived side chain [24].

Tocotrienols (vitamin E): It contains unsaturated isoprenoid side chains with three carbon-carbon double bonds. It may affect cholesterol biosynthesis by inhibiting HMG-CoA reductase [25].

Y-oryzanol: Y-oryzanol is a combination of ferulate, esters of fatty acid and triterpene alcohol which acts as an antioxidant against free radicals [26].

Phytic acid: Phytic acid is the hexaphosphoric ester of cyclohexane (inositol hexaphosphoric acid). It is usually found as a complex with essential minerals and proteins [27].

Rotary evaporator: Rotary evaporator is an instrument used to evaporate solutions in to extracts.

Methanol extract: Methanol extract is a solvent that can extract both hydrophilic and lipophilic molecules from plant parts.

Reagent: Reagent is a mixture of chemical compounds to test how a reaction occurs.

Cominution: Cominution is a reduction of solid materials from one average particle size to a smaller average particle size.

Percolation: Liquid slowly passing through a filter.

Extract: A preparation containing active ingredient of a substance in concentrated form.

Ferric reducing antioxidant power (FRAP): FRAP an assay utilized to determine the ferric to ferrous reduction of antioxidants.

Metal chelating activity: An assay performed to stimulate lipid peroxidation by Fenton reaction promoting chelation

Butylated hydroxytoluene: A standard utilized in comparing antioxidant activity or reduction.

Fenton type reaction: A chemical reaction that converts hydrogen peroxide a product of mitochondrial oxidative respiration, into a highly toxic hydroxyl free radical [28].

Chelation: Chelation is a type of bonding of ions and molecules to metal ions. It involves formation or presence of two or more separate coordinate bonds between a polydentate ligand and a single central atom (gol).

Lipid peroxidation: A free radical-mediated chain of reactions that, once initiated, results in an oxidative deterioration of polyunsaturated lipids.

REFERENCES

1. Moharram HA, Youssef MM (2014) Methods for determining the antioxidant activity: A review. Alex J Fd Sci Technol 11: 31-41.
2. Goufo P, Trindade H (2014) Rice antioxidants: Phenolic acids, flavonoids, anthocyanins, proanthocyanidins, tocopherols, tocotrienols, y-oryzanol and phytic acid. Food Sci Nutr 2: 75-104.
3. Goffman FD, Bergman CJ (2004) Rice kernel phenolic content and its relationship with antiradical efficiency. Sci Food Agric.
4. Heleno SA, Martins A, Queiroz MJRP, Ferreira ICFR (2015) Bioactivity of phenolic acids: Metabolites versus parent compounds: A review. 173: 501-513.
5. Qureshi AA, Mo H, Packer L, Peterson DM (2000) Isolation and identification of novel tocotrienols from rice bran with hypocholesterolemic, antioxidant and antitumor properties. J Agric Food Chem 48: 3130-3140.
6. Jeffrey H (2009) Rice: Its history, agriculture, production and research. Available at: http://factsanddetails.com/world/cat54/sub343/item1580.html
7. Goleniowski M, Cusido RM, Bonfill M, Palazon J (2013) Phenolic acids. In: Natural Products, pp: 1951-1973.
8. Drevon CA (1991) Absorption, transport and metabolism of vitamin E. Free Radic Res Commun 14: 229-246.
9. Brigelius-Flohé R, Traber MG (1999) Vitamin E: Function and metabolism. FASEB J 13: 1145-1155.
10. Aggrwal BB, Sundaram C, Prasad S, Kannappan R (2010) Tocotrienols, the vitamin E of the 21st century: It's potential against cancer and other chronic diseases. Biochem Pharmacol 80: 1613-1631.
11. Fu JY, Che HL, Tan DM, Teng KT (2014) Bioavailability of tocotrienols: Evidence in human studies. Nutr Metab (Lond) 11: 5.
12. Fujiwara S, Sakura S, Sugimoto I, Awata N (1983) Absorption and metabolism of gamma-oryzanol in rats. Chem Pharm Bull (Tokyo) 31: 645-652.
13. Minatel IO, Francisqueti FV, Correa CR, Lima GPP (2016) Antioxidant activity of y-oryzanol: A complex network of interactions. Int J Mol Sci 17: 1-15.
14. Viskupicova J, Ondrejovic M, Sturdik E (2008) Bioavailability and metabolism of flavonoids. J Food Nutr Res 47: 151-162.
15. Treml J, Smejkal K (2016) Flavonoids as potent scavengers of hydroxyl radicals. Compr Rev Food Sci Food Saf 15: 724-738.
16. Lila MA, Burton-Freeman B, Grace M, Kait W (2016) Unraveling anthocyanin bioavailability for human health. Annu Rev Food Sci Technol 7: 1-19.
17. Ghosh D, Konishi T (2007) Anthocyanins and anthocyanin-rich extracts: Role in diabetes and eye function. Asia Pac J Clin Nutr 16: 200-208.
18. Turrini E, Ferruzzi L, Fimognari C (2017) Possible effects of dietary anthocyanins on diabetes and insulin resistance. Curr Drug Targets 18: 629-640.
19. Graf E, Empson KL (1987) Phytic acid. J Biol Chem 262: 11647-11650.
20. Aparadh VT, Naik VV, Karadge BA (2012) Anti-oxidative properties (TPC, DPPH, FRAP, metal chelating ability, reducing power and TAC) within some cleome species. Annali Di Botanica 2: 49-56.
21. Andersen OM (2001) Anthocyanins. Encyclopedia of Life Sciences.
22. Panche AN, Diwan AD, Chandra SR (2016) Flavonoids: An overview. J Nutr Sci 5: e47.
23. Ferruzzi N (2016) Phenolic acids. Adv Food Nutr Res.
24. Velasco L, Ruiz-Mendez MV (2015) Sunflower oil minor constituents. In: Sunflower, pp: 297-329.

25. Stillwell W (2016) An introduction to biological membranes. 2nd Edn. Elsevier Science.

26. Agarwal N, Raghav PK, Sharma A (2016) Gamma oryzanol - A therapeutic agent. Int J Adv Res 4: 78-83.

27. Ahmed A, Randhawa M, Sajid MW (2014) Bioavailability of calcium, iron, and zinc in whole wheat flour. Wheat and Rice in Disease Prevention and Health, pp: 67-80.

28. Kivrak EG, Yurt KK, Kaplan AA, Alkan I, Altun G (2017) Effects of electromagnetic fields exposure on the antioxidant defense system. J Microsc Ultrastruct 5: 167-176.

29. Vinayagam R, Xu B (2015) Anti-diabetic properties of dietary flavonoids: A cellular mechanism review. Nutr Metab 12: 1-20.