PHARMACOLOGICAL ANALYSIS OF DIFFERENT VARIETIES OF RICE (*ORYZA SATIVA* LINN)

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ABSTRACT: *Oryza sativa, Asian rice, is a member of family Poaceae and main staple food for the population of several countries including Pakistan hence the phytochemical analysis of the locally consumed rice cultivars is of much importance both from commercial and consumers dietary perspectives. Keeping in view this importance eight approved varieties namely 'Sela', 'IRRI-86', 'Brown Rice', 'Superi PK-386', 'Super Basmati', 'Basmati-140', 'Basmati-150' and 'Basmati-160' of rice, were analyzed for their phytochemical analysis viz., alkaloids, saponins, coumarins, terpenoids, flavonoids, tannins, phlobatannins, cardiac glycosides case of phytochemical analysis, plant showed positive results for the presence of saponins, tannins and terpenoids by producing their characteristics precipitates, froth and ring production. Whereas, in some cases it negative results were recorded for absence of alkaloids, anthraquinones, phlobatanins, flavonoids and cardiac glycosides. The antioxidant activity revealed that the varieties of rice are rich source of antioxidants. Moreover the total antioxidant assay and DPPH assay were performed in which methanol extract of all the plant parts showed very much comparable results with the available standards as BHT and alpha tocopherol. Thus, it can be concluded that these varieties are reservoir of potentially useful chemical compounds and good source of antioxidants as well. Nonetheless, due to its dietary properties there is enormous scope of future research on different varieties of rice.*

Keywords: Rice; Poaceae; phytochemical assessment; cardiac glycosides.

1. INTRODUCTION

Rice (*Oryza sativa* L.) is a member of family *Poaceae*. It is considered the main staple food for the population of several countries, and is a major source of nutrients due to its daily consumption [1]. The first attempts to cultivate rice occurred around 10,000 years ago. Rice accounts for 21% global energy, 14% protein and 2% fat supply. Among the over 100,000 rice varieties, there are significant cultivar-specific differences in nutritional content [2]. Rice is the 2nd most important food crop in Pakistan in respect of local consumption as well as exports. It is grown on approximately more than 2.5 million hectares, with a total production of 5.0 million tons [3]. Pakistan is blessed with a multitude of agro-climatic conditions for growing several types of rice. Aromatic rice has a special place in world rice markets [4]. The best known aromatic rice includes Basmati rice. Pakistan is the producer of the world's finest long grained aromatic basmati rice. Basmati, the king of rice, is held in the highest regard world over. Aromatic (Basmati) and coarse (IRRI-6 white rice) are two main groups of rice in Pakistan [5]. The name basmati originated from a Sanskrit word "BASH", which means smell. This rice has special features, which make it naturally long grain fragrant and delicious in taste. Characteristics of basmati include aroma, fluffiness, and high kernel elongation of cooked grain. 95% of the basmati rice cultivation takes place in the province of Punjab, where total production was 2.47 million tons in 2010 [6]. Super Kernel is a long grain rice with a slender kernel, four to five times longer that it's width. The grains are separate, light and fluffy when cooked. Basmati rice 385 is dry and separate when cooked, resulting in long, thin grains, since the long grain increases only in length when cooked. Brown Rice is the least processed form of rice, as the kernels of rice have had only the hull removed. The light brown color of brown rice is caused by the presence of bran layers which are rich in minerals and vitamins, especially the B-complex group. Parboiled Rice is rough rice that has gone through a steam-pressure process before milling. It is soaked, steamed, dried, and then milled to remove the outer hull. This procedure gelatinizes the starch in the grain, and is adopted at the mill in order to harden the grain, resulting in less breakage [7]. Rice is a good source of natural antioxidants including phenolic compounds. Both the phenolic and flavonoid compounds have potential as antioxidants to act as free radical scavengers, reducing agents, and/or metal ion chelators, thus contributing to human health benefits [8]. Whilst, cereals and legumes are prominent source of antioxidants because they contain a wide array of phenolics. Phenolic acids in cereals, primarily, in bound form as conjugates with sugars, fatty acids, or proteins which act as effective natural antioxidants [9]. Therefore, the current study was conducted in order to evaluate and compare nutritional content and antioxidant properties which may give information about valuable bioactive components.

2. MATERIALS AND METHODS

The present investigation was done to check the Pharmacological potential of various varieties of *Oryza sativa*. L i.e., Sela, IRRI-86, Brown rice, Superi PK 386, Super Basmati, Basmati 140, Basmati 150, Basmati160.

The varieties of rice were collected from the locally available market. The extraction was performed by using different solvents using maceration method.Before extraction plant material was ground to make powder which was preserved in the specimen jars, until required. 10 grams of different rice varieties were extracted in sequence with different solvents. The extraction was carried out by soaking the powder in each of the solvent for the period of 8 days e.g., Petroleum ether, Chloroform and Methanol. The residue was filtered and the filtrate was preserved in the labeled glass jars, whereas the residue was further soaked in the next solvent in series.

2.1 Phytochemical Analysis

Qualitative phytochemical analysis of the extracts of the *Oryza sativa* L. was carried out by using standard procedures

to identify the constituents as described by Jamil *et al.*, 2012. Major constituents analyzed were alkaloids, tannins, terpenoids, saponins, anthraquinones, cardiac glycosides and coumarins. The results observed were based on the colour change or precipitate formation.

2.2 Antioxidant Activity

For antioxidant evaluation of the plant extracts Total antioxidant assay and DPPH radical scavenging activity was done.

2.3 Determination of Total Antioxidant Capacity

The total antioxidant capacity of all the extracts was assayed according to the method of Prieto *et al.,* 1999. 0.1ml of each solution (0.5mg/ml) was combined with 1.9ml of reagent solution (0.6M sulphuric acid, 28Mm sodium phosphate and 4mM ammonium molybdate). The reaction mixture was incubated at 95°C for 60 minutes. After cooling at room temperature, the absorbance of the mixture was measured at 695nm against a blank. The antioxidant activity will be expressed as the absorbance of the sample. The antioxidant activity of BHT (Beta hydroxyl toluine) (0.5mg/ml) was also assayed for comparison. The data was further analyzed by Analysis of Variance (ANOVA) with two ways completely randomize design to have an idea about the least significant difference among means and treatments. For results analysis Duncan's multiple range test was applied and thus the results were tabulated [10].

2.4 DPPH Radical Scavenging Activity

To evaluate the free radical scavenging activity the prepared extract of plant fractions in different solvent will be used to react with the stable radical 2,2-diphenyl-1-picrylhydrazyl radical (DPPH) according to(Erasto *et al.,* 2004). The extract solutions prepared by dissolving 0.5 mg/ml of each dry extract in respective extraction solvent that is DMSO. 0.2mg/ml DPPH in DMSO will be mixed with equal volume of test compounds, mixed well and kept in dark for 30 min. the scavenging activity on DPPH radical will be determined by measuring the absorbance at 517nm and replicate will be formed in the same manner.

3. RESULTS

Phytochemical Analysis

From the experiments the varieties contain some phytochemicals like saponins, terpenoids and tanins. Other phytochemicals like alkaloids, anthraquinones, coumarins, flavonoids, phlobatanins and cardiac glycosides were found to be absent in all varieties of rice. The results were as shown in figure 1.

3.1 Antioxidant Activity

3.1.1 Total antioxidant assay

The total antioxidant assay was performed with petroleum ether and methanol extracts to analyze them qualitatively. The results of the rice varieties extracts were being compared with standard antioxidants available (Table 1). Some of the extracts showed values very much closer to standard ones. Hence they can be used as standards. The petroleum ether extract of Basmati 150 showed the highest antioxidant value, i.e., 0.58 ± 0.015a but Brown rice and Basmati 160 in methanol extract showed value i.e., 0.53± 0.028a and 0.53± 0.015a respectively closer to the value of standard chemical α tocopherol its value is 0.513. While the petroleum ether extract of IRRI 86 and methanol extract of Superi PK 386 showed the value i.e., 0.47± 0.015c and 0.47±0.015b respectively equal to standard chemical BHT whose value is 0.476. These can be taken as standard because it closely resembles to the standard α-Tocopherol and BHT (Figure 2).

3.2 DPPH radical scavenging activity

Radial scavenging activity with DPPH Assay was performed with petroleum ether and methanol extracts to analyze them qualitatively (Table 1). The results of the plant extracts were being compared with standard antioxidants available. Extracts showed values much higher than the standard ones. Hence, they cannot be used as standards. The petroleum ether extract of Superi PK386 showed the highest antioxidant value i.e., 0.685 ± 0.172a While methanol extract of Brown rice showed highest value i.e., 0.845 ± 0.137a. The extracts showed higher values than the standards so it cannot be taken as standard because it does not resemble to the standard α-Tocopherol and BHT (Figure 2).

4. DISCUSSION

If a plant is found pharmacologically important in folk and traditional remedies, it must be evaluated through experimental studies. Pakistan is overwhelmingly an agricultural country and has vast plains irrigated by Each value is an average of three replicates ± followed by the standard deviation of each row and coriver systems. In the present research work eight approved varieties, namely Sela, IRRI-86, Brown rice, Superi PK 386, Super Basmati, Basmati 140, Basmati 150 and Basmati 160 of rice, were analyzed for phytochemicals and antioxidant attributes. Rice is one of the most important crops after wheat among cereals which is used as a source of calories and food in Pakistan; hence the phytochemical analysis of the locally consumed rice cultivars is of much importance both from commercial and consumers dietary perspectives. The phytochemical analysis of some of the locally found rice varieties was evaluated. The results revealed that all the varieties contain some phytochemicals like saponins, terpenoids and tanins. Persistant froth were produced by the plant indicated the presence of saponins. Presence of blue green ring indicated that terpenoids were present. Similarly tannins were also present by indicating brownish green coloration. These were found in small amount. Other phytochemicals like alkaloids, anthraquinones, coumarins, flavonoids, phlobatanins and cardiac glycosides were found to be absent in all varieties of rice. The results for antioxidant activity revealed that rice grain possessed good antioxidant potential. The absorption value at 695nm and

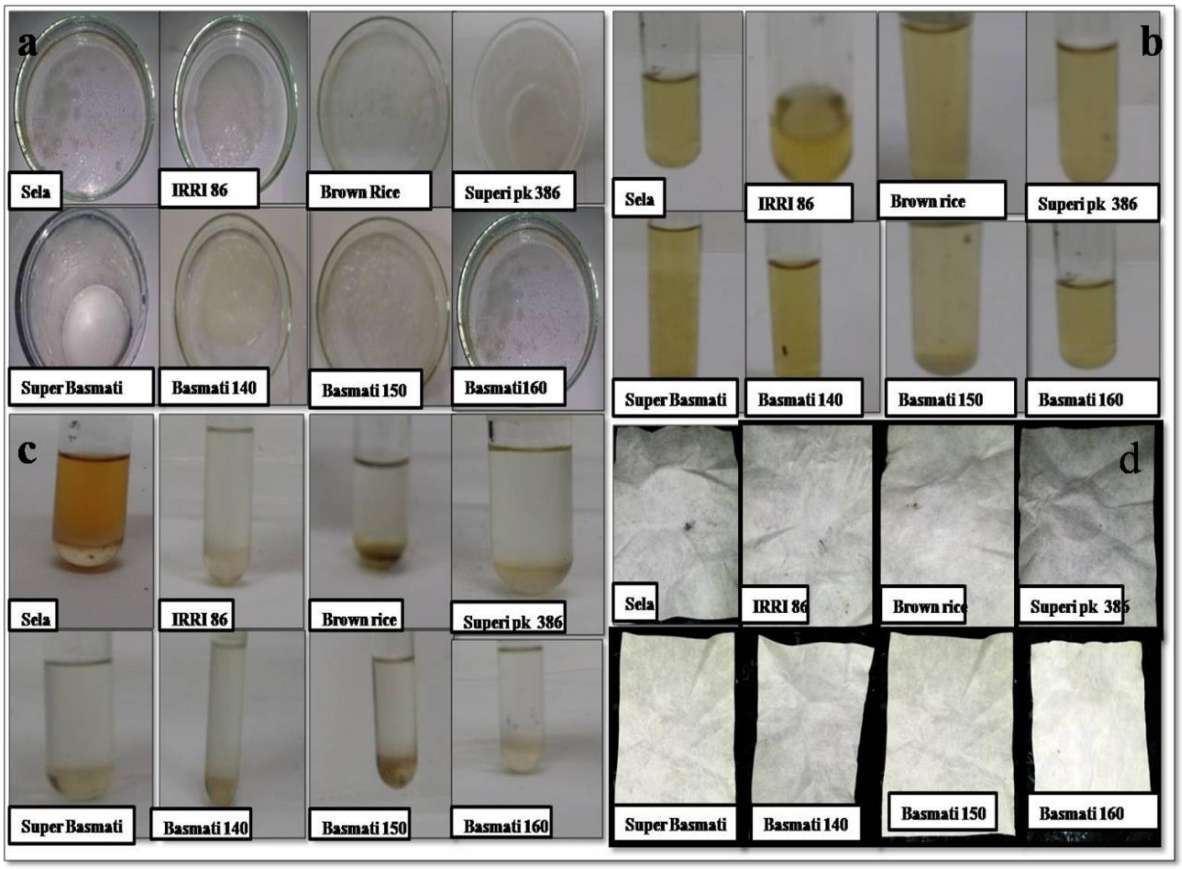


Figure 1: Indication of positive results for (a) saponins (b) tannins and (c) terpenoids whereas (d) no emission of yellow florescence in UV light thus indicating that coumarins were absent in respective samples of rice.

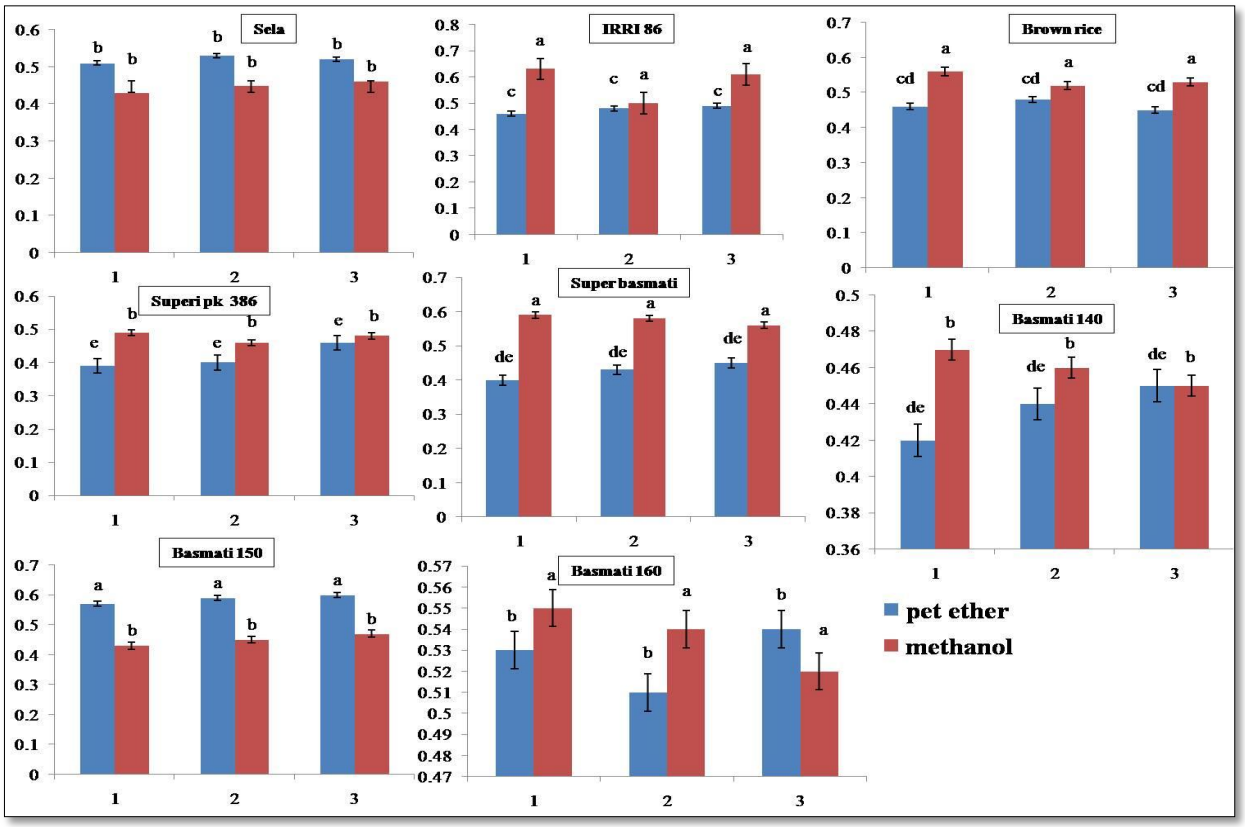
517nm of all the extracts were recorded and later on compared with the standard antioxidant chemical, BHT (Butyl Hydroxy Touline) and α-tocopherol.

Among all the varieties the absorption at 695nm, Brown rice and Basmati 160 in methanol extract showed value i.e., 0.53±0.028a and 0.53±0.015a respectively closer to the value of standard chemical α tocopherol while the petroleum ether extract of IRRI 86 and methanol extract of Superi PK 386 showed the value i.e., 0.47±0.015c and 0.47±0.015b respectively equal to

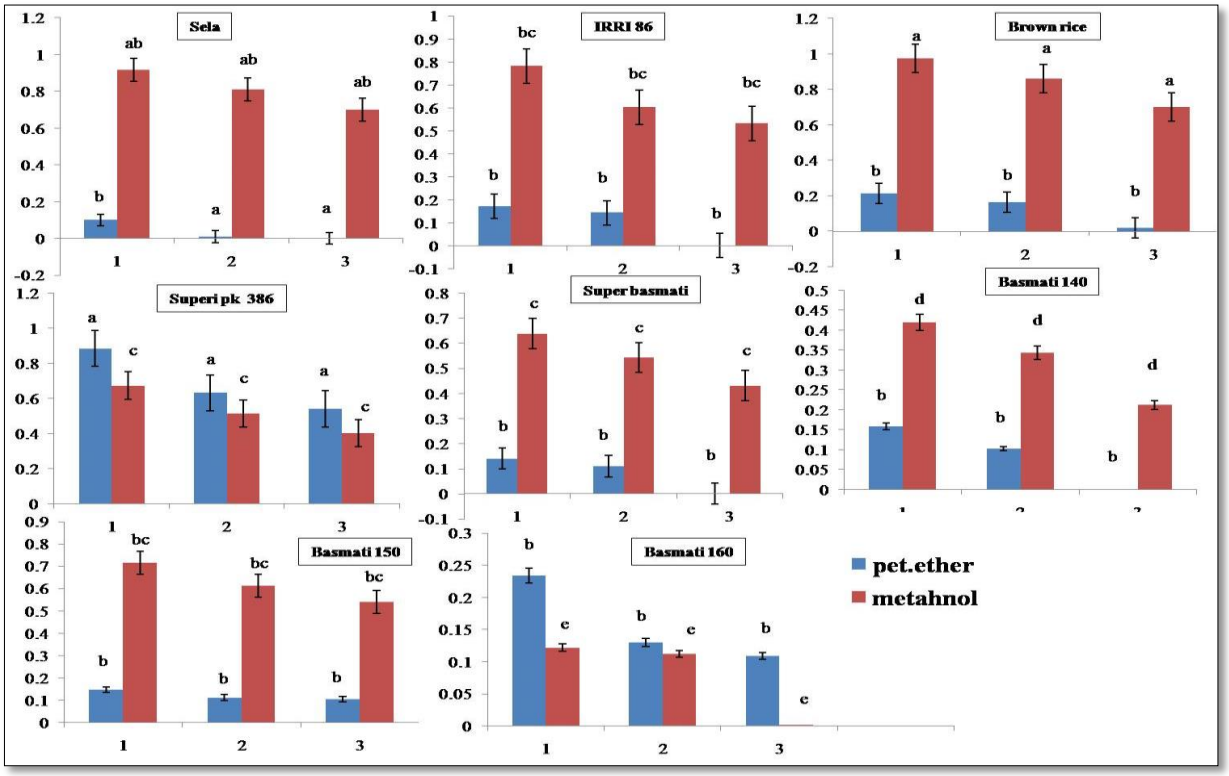
standard chemical BHT. As these values were very close to the standard samples, it means that these varieties are highly antioxidant. The absorption value at 517nm showed that the value of Superi PK 386 is higher than the standard α tocopherol *i.e.*, 0.685±0.172a. While methanol extract of Brown rice showed highest value i.e. 0.845 ± 0.137a. So these varieties did not show radical scavenging activity. The same work was done by Moongngarm *et al.*, 2012 on rice bran, rice bran layer and rice germ of four *indica* rice cultivars to determine the phytochemical and antioxidant activity. Rice germ contained highest amount of α-tocopherol, γ-tocopherol, and indicated the strongest antioxidant activity while the rice bran layer showed highest level of γ-oryzanol with the amount ranged from 5.07 mg/g in RD 6 to13.55 mg/g in black waxy rice. Chatha *et al.*, 2006 evaluated the antioxidant activity of extracts of rice bran in different solvents using different antioxidant assays. The results demonstrated that rice bran extracts of the Super Kernel variety indigenous to Pakistan are a viable source of natural antioxidants and might be exploited for functional foods and nutraceutical applications. In different studies, it has been observed that naturally occurring

Each value is an average of three replicates ± followed by the standard deviation of each row and column.

antioxidant compounds from plant sources have been identified as free radicals or active oxygen scavengers. Hence interest has been increased considerably to exploit natural resources having antioxidant activity to replace synthetic antioxidants, which are being restricted due to their side effects.



(a)



(b)

Figure 2: Antioxidant activity of extracts of eight different verities of rice in petroleum ether and methanol by (a) Total antioxidant Assay and (b) by using DPPH Assay

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Table 1: Leaf area index and plant height of 40 days old plants of 11 selected tomato genotypes at levels of stress

Genotypes	Absorption at 695 nm		Absorption at 517 nm	
	Petroleum ether	Methanol	Petroleum ether	Methanol
Sela	0.52 ± 0.01^{b}	0.44 ± 0.01^{b}	0.037 ± 0.055^{b}	0.81 ± 0.108^{ab}
IRRI 86	0.47 ± 0.015^{c}	0.58 ± 0.07^{a}	0.105 ± 0.091^{b}	0.638 ± 0.128^{bc}
Brown rice	0.46 ± 0.015^{cd}	0.53 ± 0.028^{a}	0.133 ± 0.100^{b}	0.845 ± 0.137^{a}
Superi pk 386	0.41 ± 0.037^{e}	0.47 ± 0.015^{b}	0.685 ± 0.172^{a}	0.529 ± 0.135^{c}
Super basmati	0.42 ± 0.025^{de}	0.57 ± 0.015^{a}	0.084 ± 0.072^{b}	0.537 ± 0.104^{c}
Basmati 140	0.43 ± 0.015^{de}	0.46 ± 0.01^{b}	0.087 ± 0.079^{b}	0.32 ± 0.104^{d}
Basmati 150	0.58 ± 0.015^{a}	0.45 ± 0.02^{b}	0.121 ± 0.022^{b}	0.623 ± 0.089^{bc}
Basmati 160	0.52 ± 0.015^{b}	0.53 ± 0.015^{a}	0.157 ± 0.066^{b}	0.07 ± 0.066^{e}

Standards = α- Tocopherol: 0.513; BHT: 0.476; Blank: 0.025

5. CONCLUSION

The tested all extracts of varieties of *O. sativa.* showed some of their phytochemical constituents such as saponins, terpenoids and tannins, while alkaloids, anthraquinones, coumarins, flavonoids, phlobatanins and cardiac glycosides were absent in respective plant because alkaloid, flavonoids and phenolics are present in colored varieties of rice e.g., Jyothi red color and IR64 brown color. Thus, it is concluded that these varieties are reservoir of potentially useful chemical compounds and good source of antioxidants as well. Due to its dietary properties there is enormous scope of future research on different varieties of rice.

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