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## Proximate analysis, nutritive value, total phenolic content and antioxidant activity of *Litchi Chinensis* sonn

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### ABSTRACT

*Litchi chinensis* is an evergreen tree belonging to Sapindaceae family. Lychee fruits are favored by consumers for their juicy and sweet arils, while seeds are discarded as waste material. This study was aimed to investigate the proximate analysis, nutritive value, total phenolic content and antioxidant activity of *Litchi chinensis* seed. The results of phytochemical screening revealed that seeds contain a number of medicinally active secondary metabolites. Total phenolic content was highest for ethanolic macerate (33.657 mg GAE/100 gm dw). The antioxidant activities of extracts were performed by DPPH and FRAP method. In DPPH assay the highest antioxidant activity is shown by n- butanol fraction ( $IC_{50} = 59.500 \mu\text{g/ml}$ ) while Ferric reducing antioxidant power is maximum for ethanolic macerate (707.929  $\mu\text{M/ml}$ , FRAP value =1.808) and n-butanol fraction (610.071  $\mu\text{M/ml}$ , FRAP value =1.574). The proximate analysis showed that seeds are excellent source of carbohydrate (81.098%), protein (6.126%), fat (0.891%) and crude fiber (4.327%). The nutritive value was found to be 356.917 Kcal/100 gm of seeds. Overall the results revealed that *Litchi chinensis* seeds exhibit excellent nutritive value along with potent antioxidant activity and they can be used in formulation of various pharmaceutical and cosmetic preparations against highly expensive synthetic antioxidant. © 2013 Trade Science Inc. - INDIA

### KEYWORDS

Litchi chinensis seeds;  
Maceration;  
Phytochemical constituent;  
Proximate analysis;  
Nutritive value;  
Total phenolic content;  
Antioxidant activity.

### INTRODUCTION

Since the beginning of human civilization, medicinal plants have been used by mankind for its therapeutic value. Nature has been a source of medicinal agents for thousands of years and an impressive number of modern drugs have been isolated from natural sources. Many of these isolations were based on the uses of the agents in traditional medicine. Traditional medicines include herbal, ayurveda, siddha, unani, and Chinese

medicine system. The herbal medicines provided by plants are used in form of phytochemicals, which can be defined in the strictest sense, as chemicals produced by plants. However, the term is generally used to describe chemicals from plants that may affect health, but are not essential nutrients. While there is ample evidence to support the health benefits of diets rich in fruits, vegetables, legumes, whole grains and nuts, evidence that these effects are due to specific nutrients or phytochemicals is limited. Because plant based foods

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are complex mixtures of bioactive compounds, information on the potential health effects of individual phytochemicals is linked to information on the health effects of foods that contain those phytochemicals<sup>[1]</sup>.

In the last few decades, natural fruits, fruit extracts and seeds have received much attention as sources of bioactive substances such as antioxidants, antimutagens and anticarcinogens<sup>[2,3]</sup>. One of such natural source is Lychee. *Litchi chinensis* is an evergreen tree belonging to the Sapindaceae family is widely cultivated in subtropical areas for fruits<sup>[4]</sup>. Litchi is considered as one of the best fruits due to its high nutritive value, sweet-acidic taste, excellent aroma, and bright red colour of its peel. Lychee fruits are favored by consumers for their juicy and sweet arils and attractive red pericarp<sup>[5]</sup>. Medicinally fruit of lychee is tonic to heart, brain, and liver; allays thirst; very wholesome to the body. The root bark and flower are used in form of decoction as gargle for throat infections. The seeds in Malay Peninsula are used by Chinese as anodyne and are prescribed in various neuralgic disorders and in orchitis<sup>[6]</sup>.

A very negligible work on antioxidant activity of *Litchi chinensis* seeds has so far been reported, so the present study is designed to evaluate the antioxidant nature of *Litchi chinensis* seeds, its proximate analysis and nutritive value. Phytochemical screening and total phenolic content is performed in order to establish a correlation between bioactive constituents and biological activity. Results of this study will help to provide a better understanding of natural antioxidants and further exploration can give rise to a new lead molecule which could be cheaper antioxidant in the field of cosmetic, pharmaceutical and nutraceuticals.

## EXPERIMENTAL

### Plant material

*Litchi chinensis* fruits were collected from Dehradun, Uttarakhand (India) in the month of June and duly identified and authenticated by Botanical Survey of India, (BSI) Dehradun with accession No. 113638. A voucher specimen has been deposited in medicinal plants herbarium in Department of Chemistry, Gurukula Kangri Vishwavidyalaya under the registry No. 23/15. The collected fruits were harvested for seeds, washed, dried in shade and finally grinded to

powdered form and stored in polythene bags for further use.

### Chemicals and reagents

(TPTZ) 2,4,6-tri-(2-pyridyl)-1,3,5-triazine (Hi-media), (DPPH) 1,1-diphenyl-2-picrylhydrazyl (Sigma) Folin's ciocalteau phenol reagent (Merck), Gallic Acid (Loba chemie), Ascorbic acid (Merck), Petroleum ether (Merck), n-Butanol (CDH), Methanol (Merck) were purchased. All the other solvents and chemical used were of HPLC or analytical grade.

### Instruments

Conical flask 2000 ml (Merck), Rotary vacuum evaporator (LABCO), Kjeldahl flask (JSGW), UV-Visible Spectrophotometer-2500 (Shimadzu).

### Physical evaluation

The powdered seeds were processed to undergo physical evaluation where ash value and extractive values were determined. Ash value includes the estimation of total ash, acid insoluble ash, acid soluble ash, water soluble ash and sulphated ash, while the extractive value involves the estimation of alcohol soluble and water soluble extractive value. All the parameters were performed according to standard pharmacopoeial methods<sup>[7]</sup>.

### Proximate analysis

Proximate analysis of the seeds includes the estimation of moisture content<sup>[8]</sup>, total nitrogen content, crude protein<sup>[9,10]</sup>, crude fat<sup>[11,12]</sup>, crude fiber<sup>[13]</sup>, total carbohydrate content and available carbohydrate content. While the total carbohydrate content was estimated by the difference of mean values: 100 - (% of ash + % of moisture + % of crude fat + % of crude protein) and available carbohydrate content by the difference of (% Total Carbohydrate Content - % Crude fiber).

### Nutritive value of seeds

Nutritive value of the seeds was calculated on the basis of data of proximate analysis using general Atwater factors with the help of equation<sup>[14]</sup>.

$$\text{Nutritive value (Kcal/100 gm)} = (4 \times \% \text{ Protein}) + (9 \times \% \text{ Crude Fat}) + (4 \times \% \text{ Carbohydrate})$$

### Extraction of plant material

The powdered seeds are extracted by maceration

followed by fractionation. Briefly 855 gm of powdered seeds were loaded in 2000 ml conical flask and soaked in ethanol (1500ml x 2) at room temperature (30-35°C) for 6 days each time. Concentration of the macerated solution under vacuum in a rotary evaporator gives a reddish brown semisolid (42.351 gm, 4.593%), most of which (40.500 gm) was dissolved in 200 ml of double distilled water (DDW) and then fractioned sequentially with petroleum ether (150 ml x 4) and n - butanol (150 ml x 4). The fractioned solutions were concentrated at reduced pressure in a rotary evaporator in order to minimize the loss of thermo labile compounds.

### Phytochemical screening

Phytochemical analysis for various phytoconstituents of the obtained seed extracts was undertaken using standard qualitative methods<sup>[15,16]</sup>. The obtained extracts were screened for the presence of biologically active compounds like alkaloids, carbohydrates, glycosides, inulin, protein, amino acid, Steroids and triterpenoids, fats and oil, tannins and Phenolic group, flavonoids, gums and mucilage and naphthoquinones.

### Determination of total phenolic content

The total phenolic content of the obtained extracts of *Litchi chinensis* seeds was determined with Folin-Ciocalteu assay according to the method of Singleton and Rossi with a little modification<sup>[17]</sup>. Briefly an aliquot (1 ml) of extracts (1000 µg/ml) or standard solution of gallic acid (100 - 700 µg/ml) was added to a 50 ml volumetric flask, containing 35 ml of distilled water. A blank of distilled water was also prepared. 2.5 ml of Folin-Ciocalteu's phenol reagent was added to the mixture and shaken. After 8 min of incubation at room temperature, 7.5 ml of 20 % Na<sub>2</sub>CO<sub>3</sub> solution was added to the mixture. The solution was diluted to 50 ml with distilled water and mixed. After incubation for 2 hrs at room temperature, the absorbance was measured at 765 nm with an UV- VIS spectrophotometer. Calculations were performed using the calibration curve of gallic acid. The total phenolic content of seed extracts were expressed as milligrams of gallic acid equivalents (GAE) per 100 grams dry mass (mg GAE/100 g dw). All the samples were analyzed in duplicates.

### DPPH free radical scavenging assay

The free radical scavenging assay of all the obtained

extracts of *Litchi chinensis* seeds was evaluated by stable DPPH free radical according to the method of Brand-Williams et. al with some modification<sup>[18]</sup>. A working solution of 0.004% was freshly prepared by dissolving 10 mg of DPPH in 250 ml of methanol. 1 ml of each solution of different concentration (1, 5, 10, 50, 100, 500 µg/ml) of each extract was added to 3 ml working solution of DPPH. After 30 min the absorbance of the preparations were taken at 517 nm with an UV- VIS spectrophotometer which was compared with the corresponding absorbance of standard ascorbic acid of similar concentrations (1-500 µg/ml). 1 ml of methanol with 3 ml of working DPPH solution serves as blank. Then the % radical scavenging activity or % inhibition or % reduction in color or % antiradical activity was calculated by equation:

$$\% \text{ Inhibition} = \frac{\text{(Absorbance of blank)} - \text{absorbance of sample}}{\text{(Absorbance of blank) after 30 min}} \times 100$$

IC<sub>50</sub> of the all extracts and standard ascorbic acid was calculated by graphical method by plotting % inhibition vs concentration.

### Ferric reducing antioxidant potential assay (FRAP)

The FRAP assay was done according to Benzie and Strain with some modification<sup>[19]</sup>. The stock solutions includes 300 mM acetate buffer (3.1g Sodium acetate trihydrate and 16 ml Glacial acetic acid), pH 3.6, 10 mM TPTZ (2,4,6-tri-(2-pyridyl)-1,3,5-triazine) solution in 40 mM HCl, and 20 mM FeCl<sub>3</sub>.6H<sub>2</sub>O solution. The working FRAP reagent was freshly prepared by mixing acetate buffer, TPTZ solution and FeCl<sub>3</sub>.6H<sub>2</sub>O solution in proportion of 10:1:1 (v/v) and then warmed at 37°C before using it. Antioxidant potential was determined by reacting a mixture 1 ml of each extracts (500 µg/ml) and 10 ml of working FRAP reagent. Absorbance of colored product (ferrous tripyridyltriazine complex) was then taken at 593 nm after 15 min of incubation at 37°C. Ascorbic acid standard solutions were tested in a similar way. The standard curve was linear between 100 - 800 µM ascorbic acid. Working FRAP reagent serves as blank and 1 ml

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of methanol with 10 ml of working FRAP reagent act as control. Calculations were made by calibration curve. Results were expressed as  $\mu\text{M/ml}$ . Additional dilution was needed if the FRAP value measured was over the linear range of the standard curve. FRAP value of sample was calculated by equation:

$$\text{FRAP value of Sample} = \frac{\text{(Change in absorbance of Sample from 0 to 15 minute)} \times \text{FRAP value of standard from 0 to 15 minute}}{\text{(Change in absorbance of Standard from 0 to 15 minute)}}$$

### Statistical analysis

Experimental results are given as mean  $\pm$  standard deviation (SD) of triplicate measurements. The data were statistically analyzed using the statistical program (sigmastat ver. 2.0.) The significant differences between means were calculated by a one way analysis of variance (ANOVA) using dunnett multiple-range test at  $P < 0.05$ .

## RESULTS AND DISCUSSION

### Physical evaluation

The results of physical evaluation of *Litchi chinensis* seeds are shown in TABLE 1. The result shows that seeds contained 2.164% total ash. Total ash value is a diagnostic purity index. It represents the physiological and non-physiological ash. The physiological ash gets dissolved in the dilute acid while some of the non-physiological ash remains undissolved<sup>[20]</sup>. Seeds contain 1.864% acid soluble ash and 0.299% acid insoluble ash. The acid-insoluble ash value measures the amount of silica, especially siliceous earth, present in the drug plant<sup>[21]</sup>. The sulphated ash 2.652% in seeds gives a more reliable ash figure for sample containing varying amount of volatile inorganic substances that may be lost at the ignition temperature used. It also gives an estimate of the lignin content that remains after treatment with concentrated sulphuric acid<sup>[22]</sup>. Extractive value shows 7.408% alcohol soluble and 11.437% water soluble extractive content. The extractive value indicates that for *Litchi chinensis* seeds, water is the best solvent when extraction is exhaustive.

TABLE 1 : Physical evaluation of *Litchi chinensis* seeds.

Parameter	Sub-Parameter	Results (%)*
Ash Value	Total Ash value	2.164 $\pm$ 0.12
	Acid Insoluble Ash value	0.299 $\pm$ 0.02
	Acid Soluble Ash value	1.864 $\pm$ 0.10
	Water Soluble Ash value	1.494 $\pm$ 0.75
Extractive Value	Sulphated Ash value	2.652 $\pm$ 0.77
	Ethanol soluble extractive value	7.408 $\pm$ 0.62
	Water soluble extractive value	11.437 $\pm$ 0.29

\*Results are expressed as mean of 3 values  $\pm$  standard deviation.

### Proximate analysis

The results of proximate analysis of *Litchi chinensis* seeds are shown in TABLE 2. The proximate analysis revealed moisture content of 9.721%, which is within the acceptable limits of about 6 to 15% for most vegetable drugs<sup>[21]</sup>. Low moisture content reduces errors in the estimation of the actual weight of drug material, reduces components hydrolysis by reducing the activities of hydrolytic enzymes which may destroy the active components, and also reduces the proliferation of microbial colonies and therefore minimize the chance of spoilage due to microbial attack<sup>[20]</sup>. The result shows that seeds of *Litchi chinensis* are good source of protein, fat and crude fiber. The appropriate fiber content 4.327% shows its dietary importance because high fiber contents increases digestibility. The high carbohydrate content 81.098% is beneficial since carbohydrate constitutes a major class of naturally occurring organic compounds that are essential for the maintenance of plant and animal life and also provide raw materials for many industries<sup>[23]</sup>.

TABLE 2 : Proximate analysis of *Litchi chinensis* seeds.

Parameter	Results (%)*
Moisture Content	9.721 $\pm$ 0.11
Total nitrogen Content	0.980 $\pm$ 0.52
Crude Protein	6.126 $\pm$ 0.77
Crude Fat	0.891 $\pm$ 0.09
Crude Fiber	4.327 $\pm$ 0.87
Total Carbohydrate Content	81.098 $\pm$ 0.26
Available Carbohydrate Content	76.771 $\pm$ 0.26

\*Results are expressed as mean of 3 values  $\pm$  standard deviation.

### Nutritive value of seeds

The nutritive value of seeds as calculated using gen-



eral Atwater factors with the help of above mentioned equation and it is found to be 356.917 Kcal/100 gm of seeds. The high nutritive value suggests that seeds can be used as formulation in various dietary supplements and can be a source of feed and fodder for animals.

### Extractive yield

The results for extractive yield, appearance and consistency of different extract of *Litchi chinensis* seeds are shown in TABLE 3. The yield of ethanol extract is expressed with respect to the material kept for maceration while the yields of fractions are expressed with reference to the ethanol extract (40.500 gm) dissolved in double distilled water. The yield is maximum in n-butanol fraction followed by petroleum ether and least in the remaining aqueous fraction.

**TABLE 3 : Extractive yield of different extracts of *Litchi chinensis* seeds obtained by maceration followed by fractionation.**

Extract / Fractions	Appearance	Consistency	% Yield (w/w)
Ethanol extract	Reddish brown	Semisolid	4.593*
Aqueous fraction	Transparent red	Solid	17.774**
Petroleum ether fraction	Brown	waxy	18.733**
n- Butanol fraction	Pale reddish brown	Sticky semisolid	24.687**

\*Extractive yield with respect to material, \*\* Extractive yield with respect to ethanol extract (40.500gm).

### Phytochemical screening

The results for phytochemical screening of *Litchi chinensis* seeds extracts are shown in TABLE 4. The result of phytochemical screening reveals that *Litchi chinensis* seeds contain a number of secondary metabolites like alkaloids, carbohydrates, glycosides, inulin, protein, amino acid, Steroids, triterpenoids, fixed oils, fats, Phenolic group, and flavonoids. The presence of these phytoconstituents suggests that the seeds of this plant might be of medicinal importance to pharmaceutical industries. Seeds are devoid of gums, mucilage and naphthoquinones. Phytochemical screening of the extracts shows that beside ethanol maceration maximum phytoconstituents are present in n-butanol fraction to which the extractive yield is also suggestive. In general phytoconstituents are much more exposed and extracted well in cold rather than in hot extraction; it may be due to the thermo labile nature of some phytoconstituents on exposure to high temperature.

According to Varadarajan et al.<sup>[24]</sup> the secondary metabolites and other chemical constituents of medicinal plants account for their medicinal value.

**TABLE 4: Phytochemical constituents in *Litchi chinensis* seedsextract/fractions.**

Phytoconstituents and Test performed	Extracts				
	ET	AQ	PE	BT	
Alkaloids	Mayer's Test	+	-	-	+
	Wagner's Test	-	-	-	+
	Hager's Test	+	+	+	+
	Tannic acid Test	-	-	-	+
	Molisch's Test	+	+	-	+
Carbohydrate	Fehling's Test	+	+	-	+
	Benedict's Test	+	+	-	+
	Selivanoff's Test	+	+	-	+
Anthraquinone glycosides	Borntrager's Test	-	-	-	-
	Hydroxyanthraquinones	+	+	-	+
	Keller-Killiani Test	+	-	+	+
Glycosides	Cardiac glycosides	+	-	+	+
	Legal's Test	+	-	+	+
	Baljet's Test	-	-	-	-
Saponin glycosides	Froth formation Test	+	+	-	+
	Mg and HCl reduction	+	+	+	+
Inulin		+	+	+	+
	Heat Test	+	+	-	-
Protein	Biuret Test	+	+	-	+
	Xanthoproteic Test	+	+	-	+
Amino Acid	Ninhydrin Test	+	+	-	-
Steroids/ Triterpenoids	Salkowski Test	+	-	+(T)	+
	Spot Test	+	-	+	-
Fixed oils and Fats	Saponification Test	+	-	+	-
	Shinoda Test	+	-	+	+
	Alkaline reagent Test	+	-	+	+
Flavonoids	Zinc hydrochloride Test	+	-	+	+
	Lead Acetate Test	+	+	-	+
	Ferric chloride Test	+	+	-	+
Phenolic compounds and Tannins	Test for Catechin	-	-	-	-
	Test for Chlorogenic acid	+	-	-	-
Gums and Mucilage		-	-	-	-
	Juglone Test	-	-	-	-
Naphthoquinone	Dam-Karrer Test	-	-	-	-

+ : Present; - : Absent; ET: Ethanol; AQ: Aqueous; PE: Petroleum ether; BT: n – Butanol; (T): Triterpenoids; (S): Steroids.

### Total phenolic content

The total phenolic content of *Litchi chinensis* seed

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extracts was determined by Folin-Ciocalteu method. The calibration curve for gallic acid is shown in Figure 1. The total phenolic content was expressed as mg GAE/100 g dw using the standard curve equation:  $y = 0.0014x + 0.0278$ ,  $R^2 = 0.9983$  where  $y$  is the absorbance at 765 nm and  $x$  is the total phenolic content in 1000  $\mu\text{g/ml}$  of extract. The result for total phenolic content is shown in TABLE 5. The total phenolic content was found to be maximum in n-butanol (32.657 mg GAE/100 gm dw) followed by petroleum ether (23.943 mg GAE/100 gm dw) and minimum in aqueous fraction (16.871 mg GAE/100 gm dw). Generally phenolic compounds are extracted well in the process where the temperature of solvents is not so much elevated during extraction. This may be due to the fact that phenolic compounds are susceptible towards temperature, oxygen and UV-light. Temperature may cause the loss of total phenolics due to decomposition. Light may also have a similar effect. Phenolic compounds are located in different parts of plant's tissues and cells, such as vacuoles, cell walls and cell nuclei. It is well-known that phenolic compounds contribute to quality and nutritional value in terms of modifying color, taste, aroma, and flavor and also in providing health beneficial effects. They also serve in plant defense mechanisms to counteract reactive oxygen species (ROS) in order to survive and prevent molecular damage and damage by microorganisms, insects, and herbivores<sup>[25]</sup>. Additionally, phenolics act as antimutagens and anticariogens<sup>[26]</sup>, metal chelators, antimicrobial agents and clarifying agents<sup>[27]</sup>.

**TABLE 5: Total Phenolic content of *Litchi chinensis* seed extract/fractions.**

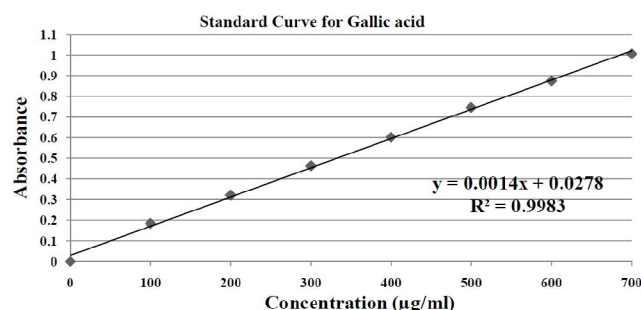
Extracts/ Fraction	Total Phenolic Content (mg GAE/100 gm dw)*
Ethanol	33.657 $\pm$ 0.96
Aqueous	16.871 $\pm$ 0.65
Petroleum ether	23.943 $\pm$ 0.88
n - Butanol	$\pm$ 1.26

\*Results are expressed as mean of 3 values  $\pm$  standard deviation.

### DPPH free radical scavenging assay

DPPH is a stable free radical which is widely used to test the free radical scavenging activities of various samples<sup>[28]</sup>. The extracts of *Litchi chinensis* seeds were subjected to screening for their possible antioxidant ac-

tivity. Antioxidant reacts with DPPH, which is a nitrogen-centered radical with a characteristic absorption at 517 nm and convert it to 1, 1-diphenyl-2-picryl hydrazine, due to their hydrogen donating ability at a very rapid rate<sup>[29]</sup>. As antioxidants donate proton to these radicals the absorption decreases. The decrease in absorption is taken as a measure of the radical scavenging. The degree of discoloration indicates the scavenging potentials of the antioxidant. The result for free radical scavenging activity of extracts/fraction is shown in TABLE 6. On comparing with standard, all fractions show excellent percentage inhibition. Figure 2 shows the graphical representation of DPPH radical scavenging assay. Figure 3 shows the graphical estimation of  $IC_{50}$ , i.e. the amount of extract as antioxidant which is able to scavenge 50% of the total DPPH radical. It was determined by drawing the graph with the sample concentration on the abscissa and the free radical inhibition (% inhibition) as the ordinate. Since  $IC_{50}$  is a measure of inhibitory concentration, a lower  $IC_{50}$  value would reflect greater antioxidant activity of the sample<sup>[30]</sup>. The antioxidant activity is maximum in n- butanol fraction having lowest  $IC_{50}$  value of 59.50  $\mu\text{g/ml}$  followed by ethanol maceration and petroleum ether fraction having  $IC_{50}$  value of 97.00  $\mu\text{g/ml}$  and 116.00  $\mu\text{g/ml}$  respectively. The DPPH radical scavenging activity of n- butanol fraction is more than that of parent ethanolic extract; this may be attributed to the fact that after fractionation some selected phytoconstituents that may be responsible for inhibition get concentrated or abstracted in n- butanol fraction where they express their antioxidant nature rather than in parent ethanolic extract. Phenolic compounds have been demonstrated to exhibit a scavenging effect for free radicals and a metal-chelating ability<sup>[31]</sup>. Ascorbic acid is a potent free radical scavenger<sup>[32,33]</sup>. So when compared to such pure component, *Litchi chinensis* seeds are potent free radical scavenger.



**Figure 1 : Callibration curve for Gallic acid.**

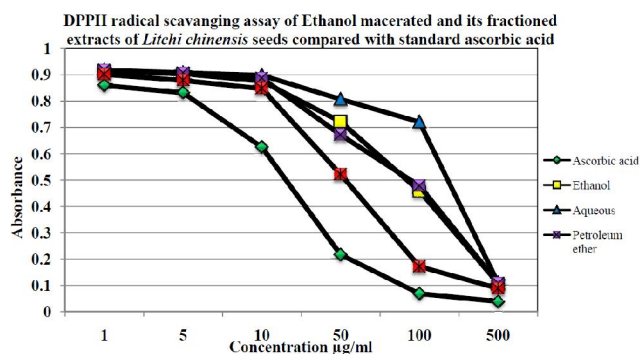


Figure 2 : DPPH radical scavenging activity of *Litchi chinensis* seeds extract/fractions.

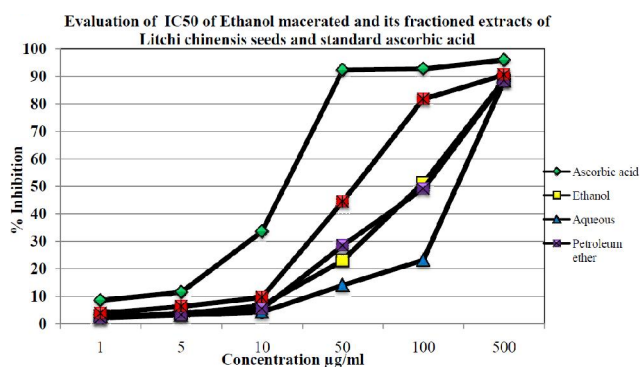


Figure 3 : Evaluation of  $IC_{50}$  of *Litchi chinensis* seed extract/fractions.

### Ferric reducing antioxidant potential assay (FRAP)

The antioxidant activity determined by FRAP method is shown in TABLE 7. Figure 4 represent the calibration curve for standard ascorbic acid. FRAP-method was initially developed to assay plasma antioxidant capacity, but can be used to measure the antioxidant capacity from a wide range of biological samples and pure compounds<sup>[34]</sup>. In this study, FRAP assay is used because it is comparatively more accurate, quick and the reaction is reproducible and linearly related to the molar concentration of the standard antioxidants. The results were expressed as  $\mu\text{M}/\text{ml}$  using the standard curve equation:  $y = 0.0014x + 0.0649$ ,  $R^2 = 0.9885$  where  $y$  is the absorbance at 593 nm and  $x$  is the ferric reducing antioxidant ability in 500  $\mu\text{g}/\text{ml}$  of extracts. The unit  $\mu\text{M}/\text{ml}$  means the quantity of  $\text{Fe}^{3+}$  in  $\mu\text{M}$  that can be reduced to  $\text{Fe}^{2+}$  by per ml of extract or standard ascorbic acid. The ferric reducing antioxidant power is maximum in parent ethanolic extract (707.929  $\mu\text{M}/\text{ml}$ ) and in n-butanol fraction (610.071  $\mu\text{M}/\text{ml}$ ) followed by aqueous (535.786  $\mu\text{M}/\text{ml}$ ) and least in petroleum ether frac-

tion (463.642  $\mu\text{M}/\text{ml}$ ). The FRAP value for ethanol macerate and n-butanol fraction is calculated to be 1.808 and 1.574 respectively which is very close to standard ascorbic acid (FRAP value = 2). FRAP method particularly helps in assessing the antioxidant behaviour of extracts in which those phytoconstituents are present which act by reducing ions or by donating electron and not by radical quenching mechanism. The FRAP activity of extracts may be due to the presence of phenolic hydroxyl or methoxyl groups, flavones hydroxyl, keto groups, free carboxylic groups and other structural features like triterpenes and their derivative<sup>[35]</sup>.

TABLE 6 : DPPH radical scavenging activity of *Litchi chinensis* seed extract/fractions.

Extract/ Standard	Concentration ( $\mu\text{g}/\text{ml}$ )	(%) Inhibition*	$IC_{50}$ value ( $\mu\text{g}/\text{ml}$ )
Ascorbic Acid	1	8.511 $\pm$ 0.15	22.72 $\pm$ 0.09
	5	11.489 $\pm$ 0.17	
	10	33.511 $\pm$ 0.22	
	50	92.234 $\pm$ 0.02	
	100	92.656 $\pm$ 0.06	
Ethanol	500	95.851 $\pm$ 0.52	97.000 $\pm$ 0.12
	1	2.772 $\pm$ 0.67	
	5	3.625 $\pm$ 0.55	
	10	6.609 $\pm$ 0.96	
	50	23.028 $\pm$ 0.23	
Aqueous	100	51.178 $\pm$ 0.17	267.000 $\pm$ 0.15
	500	88.593 $\pm$ 0.08	
	1	2.239 $\pm$ 0.62	
	5	3.198 $\pm$ 0.76	
	10	4.264 $\pm$ 0.55	
Petroleum Ether	50	13.965 $\pm$ 0.41	116.000 $\pm$ 0.29
	100	23.134 $\pm$ 0.22	
	500	88.273 $\pm$ 0.10	
	1	2.132 $\pm$ 0.89	
	5	3.305 $\pm$ 0.93	
n - Butanol	10	5.437 $\pm$ 0.96	59.500 $\pm$ 0.03
	50	28.358 $\pm$ 0.86	
	100	49.040 $\pm$ 0.99	
	500	88.380 $\pm$ 0.61	
	1	3.838 $\pm$ 0.35	
	5	6.290 $\pm$ 0.61	
	10	9.595 $\pm$ 0.39	
	50	44.349 $\pm$ 0.19	
	100	81.663 $\pm$ 0.16	
	500	90.512 $\pm$ 0.05	

\*Results are expressed as mean of 3 values  $\pm$  standard deviation.

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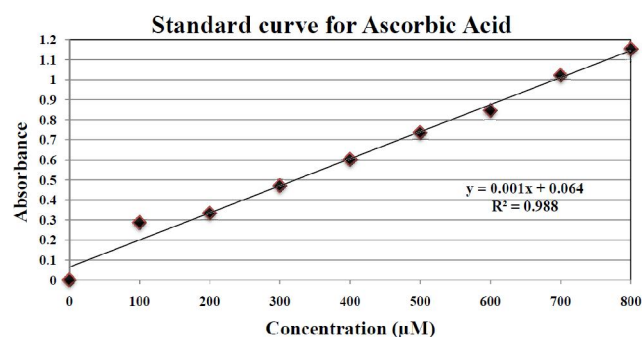


Figure 4: Calibration curve for Ascorbic acid.

TABLE 7: Ferric reducing antioxidant power of *Litchi chinensis* seed extract/fractions.

Samples	Ferric reducing antioxidant power (µM/ml)*	FRAP Value
Ascorbic acid	787.929 ± 0.14	2.000
Ethanol	707.929 ± 0.09	1.808
Aqueous	535.786 ± 1.92	1.395
Petroleum ether	463.642 ± 2.02	1.223
n - Butanol	610.071 ± 0.96	1.574

\*Results are expressed as mean of 3 values ± standard deviation.

## CONCLUSION

Lychee is one of the most popular fruit that is grown commercially for its juicy arils and nutritional benefits in various countries of world including India and China. Usually what people do, they used to consume the edible portion of the lychee fruit and discard the remaining pericarp and seeds while they are too useful. The present study aims at evaluating the nutritional benefits and antioxidant behavior of lychee seed. The results show the nutritional benefit, presence of secondary metabolites and potent antioxidant nature of lychee seeds. By this study authors strongly suggest that lychee seeds which are commonly discarded by the people can be used to abstract out a lead molecule that could act as a potent antioxidant having a number of beneficial effect over synthetic one. This will make the availability of natural antioxidant drugs to people at low cost with less or no side effect. On the other hand, in most food industries, synthetic antioxidants e.g. butylated hydroxyanisole (BHA), butylated hydroxytoluene (BHT), tertiary butyl hydroquinone (TBHQ), propyl gallate (PG) etc. are used in order to prevent the rancidity of processed foods. This experiment supports that seeds of this fruit can be used in such industries as natural antioxidants subjected to proper investigations.

The whole plant can also be further harnessed for novel antioxidant/ bioactive compounds which are very well evidenced by the present work.

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