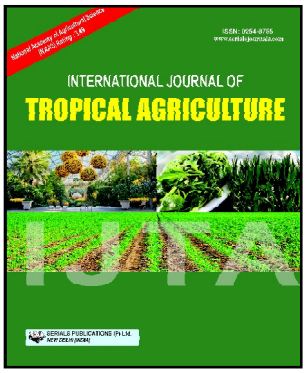

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Studies on physico-chemical properties of four local genotypes of two varieties (green and red coloured and with or without spines) of Water chestnut (*Trapa natans* L. var. bispinosa Roxb.) in Chhattisgarh

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Abstract: Water chestnut (*Trapa sp.*), locally known as singhara or paniphal is an edible aquatic dicotyledonous plant covered with a thick hard outer cover. In India it is commonly consumed after steaming once the outer cover gets soften and inside white kernel is cooked. Its kernel is a good source of protein (8.24%,) and carbohydrate (84.8%) which is usually eaten as a snack or as an ingredient in other foods and also during fasting as an ethnic food. Several varieties are found in India among which *Trapa bispinosa* Roxb. and *Trapa natans* L. are common. *Trapa bispinosa* has two varieties, one is red (Leaf, petiole and fruit) and the other is green (Leaf, petiole and fruit). Recently, the cultivation of water chestnut is becoming more popular in India due to its easy grow, low cost of production, good profit and to meet up the demand for more food production. This plant is extensively cultivated in fresh water tanks, beels, ditches, ponds and even in bunded paddy fields.

In this study, four local genotypes of two varieties (Green and red) of water chestnuts (*Trapa sp.*) (with and without spines) have been collected from the Instructional Farm Bharregaon of Pt.K.L.S College of Horticulture and Research Station, Rajnandgaon, Chhattisgarh and are selected for their physical and biochemical analysis as well using standard methods. Among the four genotypes two were of red colour (with and without thorn) and other two were green variety (with and without thorn). Of the four genotypes the most characteristic feature observed was the absence of thorns in the fruit of water chestnut. This helps the harvesting process and other cultural practices in the field condition. The physical appearance of red coloured variety was excellent. The physical characteristics like size, L/H ratio, average weight and density of the red coloured variety was found better as compared to other genotypes. The proximate composition of red coloured (without thorn) water chestnuts revealed moisture 84.2 %, ash 1.04, crude fiber 2.29%, total sugar 4.9%, reducing sugar 3.62%, non-reducing sugar 1.28%. One hundred gram of

red variety (without thorn) contained, beta-Carotene 92 microg, vitamin-C 1.10 mg and total phenol 0.62 mg. The green variety contained moisture 88.2%, ash 0.52%, crude fiber 2.13%, total sugar 3.8%, reducing sugar 2.43%, non-reducing sugar 1.37%. The green variety contained beta-Carotene 60 µg , vitamin-C 0.92 mg and total phenol 0.52 mg per 100 g. The free amino acids, glutamic acid, tryptophan, tyrosine, alanine, lysine and leucine were commonly found in both varieties. In addition, green and red variety contained cysteine, arginine and proline and glutamine and asparagines, respectively. Thus, the present study sheds light on the physic-chemical characters of the two varieties of water chestnuts and reveals that water chestnuts may play a crucial role in human nutrition.

Key words: Physico-chemical analysis, red and green genotypes, water chestnut.

INTRODUCTION

Water chestnut (*Trapa natans* L. var. *bispinosa* Roxburgh) is an edible aquatic angiosperm locally known as "Singhara" and is available in submerged low-lying areas and in village ponds. It is one of the important annual aquatic warm season crops. It is a floating (submerged rooted) plant, found commonly on the water surface of lakes, small ponds , stagnant water bodies throughout India especially in Punjab, Bihar, Uttar Pradesh, Madhya Pradesh, Chhattisgarh, Tamilnadu and Maharashtra. (Puste, 2004). Water chestnut is an important commodity in world food industry because of its unique taste (Parker and Waldron, 1995). Water chestnuts also have a firm and crispy texture, which adds to their appeal as an ingredient in meals where the vegetables to be used must have a crunchy consistency. In India the fruits of water chestnuts are eaten raw at tender stage or after boiling and roasting. It is consumed mainly in the form of boiled fruit, flour or in the shape of sweet dishes as cakes of many kinds. The dark-brown corms (whole fruit) are peeled before cooking or processing. The bulk of the edible region consists of white coloured crunchy and starch-rich, thin walled storage parenchyma. The property of firmness, crunchy texture after cooking is attributed to the lack of cell separation during cooking.

It has many medicinal values. The acrid juice is used for diarrhoea and dysentery. The fruits are used as intestinal astringent, aphrodisiac, anti-inflammatory, antileprotic, in urinary discharges, fractures, sore throat, bronchitis and anaemia.

Looking to the consumers demand for natural foods having good therapeutic values, water chestnut extends its high values in many aspects of therapeutic food. The high consumption values of the fresh fruit are probably linked to the high nutritional and organoleptic value, and also to the increasing interest of the consumers towards organic products.

The fruit has also been used in medicinal purposes (e.g., rheumatism, sunburn) are also useful in burning sensation, dyspepsia, intermittent fever fatigue, inflammation, bronchitis and debility. Flour made from fruit is suitable for textile sizing and is a good substitute for cornstarch in ice cream manufacture.

Importance of fruits in the human diet is well recognized because they are rich in essential contents like vitamins and minerals which help in the maintenance of proper health and resistance to specific diseases. Some research is going on in several research centres of our country; however, a limited work was done on the physicochemical properties of different varieties of water chestnuts. Therefore, the details research works including biochemical are very important to release a good variety of water chestnut, which would be nutritionally rich as well as high yielding. Keeping all these in mind in this study, an attempt was made to investigate in details of the water chestnut fruits using locally available

two varieties (green and red). This study comprises the determination and comparison of physiochemical characteristics Finally, the purpose of this study was to increase existing knowledge on aquatic plants by investigating the physic-chemical characteristics of the plant and to explore its suitability as a source food.

MATERIALS AND METHODS

Freshly harvested matured fruits of Water chestnut were collected from the pond of instructional farm ,Bharregaon of Pt.K.L.S.College of Horticulture and Research Station, Rajnandgaon, Chhattisgarh in the month of November-December 2015 for experimental purpose. The physical and chemical parameters were studied from January 2015 in by the following methods. The fruits were washed thoroughly with tap water; excessive water was drained and further stored at refrigerated temperature (4°C) till further use.

PREPARATION OF PLANT SAMPLES FOR ANALYSIS

Drying: A clean glass container was placed in an oven at 105°C overnight and allowed the container to cool in desiccators and weigh it. The samples were put in to the container and weight was taken. Then the container was placed in the oven at 105°C for 24 h and allowed the container to cool in a desiccator and weight was taken again. Drying cooling and weighting were repeated until the weight becomes constant. The dried sample was stored in an airtight container. The moisture content in the sample was calculated.

Grinding: The dried plant material was cut into small pieces with a knife or scissors. The sample was grinded in a plant grinder fitted with a suitable screen. When the grinding takes a long time, the sample were absorbed moisture and then it necessary to dried the sample again in the oven at 105°C overnight.

Physical Analysis

Length, width and thickness were measured at nine different points to an accuracy of 0.02 and 0.01 mm using a vernier callipers and micrometre respectively to have a mean average of all measurement.

Density was determined with 1L volumetric (10 cm dia. and 10 cm height) grain measuring cylinder. True density was measured using water displacement method with same material used for bulk density for 1L volume measuring cylinder.

Chemical Analysis

Determination of pH:

- Extraction of juice from water chestnut: About 70-90 g of water chestnuts was taken in a mortar. The fruits were crushed thoroughly in a mortar with a pestle and then filtered through two layers of muslin cloth. From the filtrate the clear supernatant was collected
- Procedure: The pH was estimated by using digital pHmeter. The equipment was raised, washed twice with distilled water, rinsed off with the juice of the cultivars and then dipped into the juice of the water chestnut. The pH of the juice was noted.Determination of Total soluble solids of the sample, the acidity, total sugars, reducing sugar of the samples were estimated as per the procedure of Ranganna (2002). The moisture and ash content were calculated using AOAC (2006) method. The moisture was converted to dry weight basis and reported as kg of moisture/ kg of dry matter.

Determination of vitamin-C: Vitamin-C content of water chestnuts was determined by the titrimetric method.).

Determination of β-Carotene: β-Carotene content of water chestnuts was determined

according to the procedure reported in methods of Biochemical Analysis (Metcalf, 1957).

Column preparation: A column was prepared by using alumina as a packing material. Ten precent acetone in petroleum ether was used as eluent buffer.

Procedure: The 5.0 g of fresh water chestnuts and about 4.0 g of ammonium sulphate were taken in a mortar and rubbed to an even paste with pestle. The extraction was carried out with acetone and small amount of hexane. Extraction was continued until the acetone extract became colorless. Potassium hydroxide solution (10 mL, 5.6%) was added to the extract and it was kept in a dark place for half an hour. The mixture was then transferred to a separating funnel, then 20 mL of petroleum ether few mL of hexane and 10 mL of water were added to the process was repeated until the petroleum ether layer became colorless. The petroleum ether, was concentrated by gentle heating, the concentrated extract (1-2 mL) was applied on to the top of the alumina column and eluted with 10% acetone in petroleum ether. The absorbance of the eluent was taken at 440 nm in a Coleman Junior II spectrophotometer.

Construction of standard curve of β-carotene: A standard curve was prepared by taking 0.0, 0.1, 0.2, 0.4, 1.6, 0.8 and 1.0 mL standard solution of β-carotene and the volume was made up to 5 mL with petroleum ether and mixed well. The absorbance of the solutions was taken at 440 nm in a Coleman Junior II spectrophotometer and a standard curve of β-carotene was prepared by plotting the data. The amount of β-carotene content in each variety of water chestnut (*Trapa* sp.) was calculated by using the standard curve.

Determination of total phenol: Total phenol content was determined colorimetrically by Folin-Ciocalteau's method.

Extraction of phenol: Extraction of phenol from Water chestnuts was done according to the methods described by Loomis and Shull.

Five to six gram of water chestnuts were cut into small pieces and immediately plunged into boiling ethyl alcohol and allowed to boil for 5-10 min. (5 to 10 mL of alcohol was used per g of fruits). The extract was cooled and crushed thoroughly in a mortar with a pestle. Then the extract filtered through two layers of muslin-cloth and re-extracted the tissue for three min. in hot 80% alcohol, using 2 to 3 ml of alcohol for each g of tissue. This second extraction ensured complete removal of alcohol soluble substances. The extract was cooled and passed through muslin cloth. Both the extracts were filtered through Whatman No. 41 filter paper. This alcohol extract was used for the estimation of total phenol.

Determination of crude fiber: Crude fiber was determined by the following method (AOAC, 1980).

RESULTS AND DISCUSSION

Physical analysis

The physical analysis of all the four genotypes of water chestnut are given in the table 1. It is clear from the data that average weight of four genotypes of whole water chestnut varies from 12.56 to 18.50 g; average volume varies from 9.80 to 16.24 ml and density in the range of 0.92 to 2.03 which is similar to the findings of Rodriguez *et al.* (1964). The water chestnut is in triangular bull's head shape varying from top to bottom wherein the average length varies from 28.02 mm to 34.23 mm. The peeled water chestnut fresh fruits dimensions are mentioned in table 1 wherein the average length 24.70 mm to 28.78 mm. Based on physical observation of four genotypes it is clear that the physical characteristics of the red coloured variety (without spine) was found better when compared to all the other three genotypes.

Chemical composition

The pH of water chestnuts was found in the acidic range of both varieties. From the results of TTA it

Table 1
Physical Characteristics of fresh whole fruit and peeled Water Chestnut kernel*

S.No.	*Parameters*	*Green colored*	*Red colored*	*Green colored with thorns*	*Red colored with thorns*
	(A) Fresh whole fruit				
01	Average Length (m.m.)	32.43	34.23	28.02	28.50
02	Average Height (m.m.)	30.22	32.26	26.52	24.82
03	Average Thickness(m.m.)	14.96	16.23	13.2	14.20
04	L/H Ratio	1.061	1.073	1.056	1.148
05	Average weight (g)	12.56	18.50	13.56	13.89
06	Average volume (ml.)	12.2	16.24	9.80	10.24
07	Density	1.03	2.03	0.92	1.01
	(B) Peeled fresh chestnut kernel				
01	Average Length (m.m.)	26.23	27.69	24.70	28.78
02	Average Height (m.m.)	23.08	25.34	21.14	26.43
03	Average Thickness(m.m.)	14.27	14.88	12.86	14.12
04	L/H Ratio	1.136	1.094	1.168	1.089
05	Average weight (g)	8.396	10.162	7.241	9.498
06	Average volume (ml.)	8.28	9.95	7.20	9.45
07	Density	1.014	1.021	1.006	0.995

*Each value represents the average value.

Table 2
Chemical composition of peeled water chestnut

S.No.	*Parameters*	*Green colored*	*Red colored*	*Green colored with thorns*	*Red colored with thorns*
01	Moisture (%)	88.2	84.2	85.2	86.2
02	Dry matter (%)	10.5	11.3	10.2	13.0
03	Crude Fibre (%)	2.13	2.29	2.0	2.10
04	pH	5.88	5.11	5.60	5.0
05	Ash (%)	0.52	1.04	0.49	092
06	TSS (%)	5.00	9.20	4.30	7.20
07	Acidity (%)	0.126	0.152	0.130	0.149
08	Ascorbic Acid (mg.)	0.92	1.10	0.90	1.0
09	Reducing Sugar (%)	2.43	3.62	3.14	3.10
10	Total Sugar (%)	3.8	4.9	4.2	4.4
11	Starch (%)	2.74	9.48	5.32	8.24
12	β-Carotene (µg)	60	92	62	90
13	Total Phenol (mg)	0.52	0.62	0.52	0.60

*Each value represents the average value.

was observed that the acidity of the water chestnuts was decreased in green variety. The moisture contents were found to be in the range of 84.2 % and 88.2% in the red and green varieties of water chestnuts, respectively.

Dry matter content analysis revealed a marked increase of dry matter as derived from percentage of moisture content in red variety as compared to green variety. The increased in dry matter content showed good correlation with the decrease in moisture content.The ash content was observed 0.52 and 1.04% in green and red varieties, respectively.

The amounts of crude fiber in two cultivars of the water chestnuts were determined to be 2.13 and 2.29% for green and red varieties, respectively.

The amount of total sugar was estimated to be 3.8% (green) and 4.9% (red). It was found that water chestnuts contained low amount of reducing sugar and the content of reducing sugar was estimated to be 2.43 and 3.62% in green and red varieties, respectively. It is reported that in case of water chestnuts the reducing sugar content is affected by several factors including variety, growing conditions, maturity and the storage environment (Mazumdar, 1985).

In contrast to the above findings, recent research has emphasized the role of ascorbic acid in the conversion of folic acid to a physiologically active form tetrahydrofolic acid. It may be concluded from the results that the vitamin-C content of water chestnuts

Table 3
Amino acid content of the four genotypes of water chestnuts

S.No.	*Amino Acid*	*Green colored (without spines)*	*Red colored (without spines)*	*Green colored with spines*	*Red colored with spines*
01	Alanine	+	+	+	+
02	Arginine	+	-	+	-
03	Aspartic acid	-	-	-	-
04	Asparagine	-	+	-	+
05	Cystine	+	-	+	-
06	Valine	-	-	-	-
07	Glutamine	-	+	-	+
08	Glutamic acid	+	+	+	+
09	Histidine	-	-	-	-
10	Hydroxyproline	-	-	-	-
11	Isoleucine	-	-	-	-
12	Leucine	+	+	+	+
13	Lysine	+	+	+	+
14	Methionine	-	-	-	-
15	Phenylalanine	-	-	-	-
16	Proline	+	-	+	-
17	Serine	-	-	-	-
18	Threonine	-	-	-	-
19	Tyrosine	+	+	+	+
20	Tryptophan	+	+	+	+

*(+, - sign represents the presence and absence of amino acid respectively)

was higher (1.10 mg/100 g) in red variety compared to green variety (0.92 mg/100 g). These results suggest that water chestnuts are a good source of vitamin-C.

β-Carotenes are known as precursors of vitamin A. In plants, it is required for growth and development of soft tissues through its effect upon protein synthesis. Therefore, in the present study, importance is also given to measure the β-Carotene content of different varieties of water chestnuts The data presented in table 2 indicates that β-Carotene content was higher in red variety (92 µg) and lower in green variety (60 µg).

Phenolic compounds are playing an important role in determining color and flavor (Lee and Jaworski, 1987). Water chestnuts contained low amount of phenol. In green variety the amount of phenol was 0.52 mg (per 100 g) and in red varieties the amount of phenol was 0.62 mg (per 100 g). It may be concluded from the results that the phenol content of water chestnuts was slightly higher in red variety. These results are indirectly supported by a report of Malviya *et al.* (2010).

The free amino acids present in the different varieties of water chestnuts was identified for the first time by two dimensional paper chromatography. As shown in Table 3, nine amino acids were detected in the green variety whereas, eight amino acids were detected in the red variety. The amino acids, glutamic acid, tryptophan, tyrosine, alanine, lysine and leucine were present in both of the varieties. Moreover, cysteine, arginine and proline were detected only in the green variety. Furthermore, the red variety but not the green variety contained free amino acids, glutamine and asparagine.

CONCLUSION

From this study, it appears that there are very few differences in physicochemical properties and nutrients composition between the two varieties of water chestnuts. However, only a significant difference was observed in the case of the â-carotene, and some amino acids contents in the both of the varieties of water chestnuts. The present study has demonstrated that, Water chestnuts which is consumed by the people either fresh or cooked may be an important source of nutrients and thus, suggesting its suitability for incorporation in human diet. Moreover, this biochemical analysis may contribute a great in supplementing existing knowledge on aquatic plants particularly on the physico-chemical study of the plant.

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