

Nutritive values, dietary antioxidant and seed protein profile of some under-utilized seeds and nuts from ethnic sources

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Received: January 2019 / Accepted: January 2019

Abstract

Study with seven unrelated and diverse type of non-conventional seeds and nuts, viz. *Trapa natans*, *Nymphaea nouchali*, *Euryle ferox*, *Castanopsis argentea*, *Artocarpus heterophyllus*, *Nelumbo nucifera* and *Gnetum gnemon* revealed that they are rich in nutritive values and high in calorific values. Crude protein content varied from 4.40-32.08%; carbohydrate content varied from 38.66-77.50%. With calorific values in the range of 317.78 Kcal/100g to 376.62 Kcal/100g, these are high calorie food with low lipid in the range of 0.93% to 6.74%. They are also rich in dietary antioxidant phenolics which varied widely from 1.0-17.6 mg/g. The free amino acid content varied from 4.0-10.66 mg/g. Present study show that that non-conventional seeds and nuts are very rich in nutrient content

Key words: Nutritive value, seeds and nuts, phenolics, non-conventional food plants

Introduction

Edible non-conventional seeds and nuts and their consumption is a centuries old ethnic practice in different tribal areas as well as among rural communities in India and many other countries. These are mostly seasonal supplementary food, while some others are popular delicacy and some others are used as scarcity food at time of famine (Arora and Pandey 1996, Singh et al. 2013). Some non-conventional seeds and nuts like Jackfruit (*Artocarpus heterophyllus*), Lotus (*Nelumbo nucifera*), Makhana (*Euryle ferox*), Chestnut (*Castanopsis argentea*), Water chestnut (*Trapa natans* var. *bispinosa*), Mokuia (*Nymphaea nouchali*), Gnetum (*Gnetumgnemon*), etc are sold in rural as well as urban market as low cost food item. This is an indication of their consumer acceptance and popularity. Unlike conventional food plants like paddy, wheat, pulses etc they do not come from organized cultivation; rather they are collected from wild, semi-wild habitat, while few are grown in backyard of rural household and hence considered as non-conventional food plants. Although they are part of traditional knowledge and ethnic culture, yet scientific scrutiny about them are scarce. In the absence of scientific scrutiny there is a general tendency to look them down as nutritionally poor and unimportant

with little or no contribution to food security system. In the backdrop of growing awareness about bio-resources and growing fear of food crisis, it is important to assess such little known non-conventional seeds and nuts for their nutritive and other values including characterization. This is likely to widen our food base and contribute to our food basket.

Materials and Methods

Non-conventional seeds and nuts of seven different and diverse plant species were collected from their natural habitat for the present study. These are– *Trapa natans* (Water chestnut or Pani singhara, Trapaceae)- a free floating aquatic herb with blackish or purple coloured triangular nut with soft, white, fleshy kernel inside. *Nymphaea nouchali* (Mokuia, Nymphaeaceae)- an aquatic herb with globose fruit that contain numerous blackish seeds embedded within pulpy mesocarp that constitute the edible part. *Castanopsis argentea* (Chest nut, Fagaceae)- a tall tree growing at an altitude of about 5000 ft. in Meghalaya and Nagaland. The nuts, little bigger than pea, is covered with soft spine at growing stage but fall off after maturity. The local tribals roast the mature nuts following which it become completely free of the spiny structures and the surface become smooth. It is popularly referred to as groundnut substitute for its similarity in taste with groundnut. *Artocarpus heterophyllus* (Jackfruit, Moraceae) is an evergreen tall

tree. Jackfruit seeds are consumed as delicacy and as potato substitute. *Nelumbo nucifera* (Lotus, Nymphaeaceae)– well known aquatic plant, known as India's national flower. *Gnetum gnemon* (Gnetum, Gnetaceae)– a rare type of gymnosperm and perennial shrub that grow as undergrowth in forest. All these seeds and nuts are sold in local market and road side. Moreover, for the present study they were collected from their natural habitat.

Nutritive values were analysed in terms of major nutritional components (AOAC 1975). Freshly collected seeds or nuts were manually dehusked, cleaned and cut into fine pieces. The samples were dried overnight at room temperature and then dried in hot air oven at a constant temperature of 60 °C till constant weight was recorded. The dried samples were grounded into fine powder in a mortar and pestle. Crude protein was estimated by microkjeldahl method (Bagchi et al. 2004). Total carbohydrate was estimated by anthrone method outlined by Clegg (1956). Total soluble sugar from the sample was extracted with warm 80% ethanol. Subsequently ethanol was removed by evaporation and the residual extract was dissolved in distilled water. Estimation was made as per the anthrone method. Lipid content was determined by extracting the sample with petroleum ether for eight hours in Soxhlet apparatus and then removing the solvent by fractional distillation (Bagchi et al. 2004). Crude fibre in the sample was determined as per the protocol of Sadasivam and Manickam (1992). Total mineral in the form of ash content was determined by ashing the sample at 600°C for three hours (Bagchi et al. 2004). The data were recorded as percentage of dry weight and calorific values were computed using the formula of Sherman (1952). Free amino acids were extracted with 80% warmethanol and quantified by spectroscopic method (Gopalan et al. 1995). Total phenolics were estimated spectroscopically using Folin-Ciocalteu as chromogenic reagent and gallic acid as standard (Gopalan et al. 1995). Seed protein profile was analysed by the standard SDS-PAGE technique outlined by Laemmli⁷ using 12% separating gel and 4% stacking gel. For Lotus, Jackfruit and Water-chestnut seed protein was extracted with 0.3M tris-HCL (pH 6.5) and for the rest 0.2M phosphate buffer (pH 7.5) was used as extraction buffer. The extraction buffers were selected by trial and error process. For determination of molecular weight, standard protein maker (PMW-M, Bangalore Genei) was co-electrophoresed.

Results and Discussion

Wide variation has been observed for protein content among the species *T. natans* which is a popular delicacy

and fetch reasonable price in the local market has 10.04% protein which is reasonable. Jack fruit seed is traditionally used as potato substitute or potato equivalent and it has 15.0% protein which is far more superior to potato which has only 4.0 – 6.3% protein(8). Nuts of *C. argentea* and seeds *G. gnemon* are popular in hilly states of North-East India as a delicacy like roasted groundnuts. While *C. argentea* nut has poor level of protein with 4.4%, *G. gnemon* has a reasonable amount of proteins with 14.78%. But lotus seeds are most outstanding with 32.08% which is higher than any pulse grain except Soyabean. It is noteworthy that except *T. natans* and *N. nouchlai* seeds and nuts can be stored for long time with proper drying which brighten the prospect of organized cultivation of those underutilized seeds and nuts. All the seeds and nuts can be eaten raw or after cooking or roasting except Jackfruit whose seeds are consumed only after cooking. Like protein, total carbohydrate and total soluble sugar also exhibited wide variation. Among them *T.natans* is characterized by high level of total carbohydrate (70%) and as well as total soluble sugar (6.0%). Soluble sugar mostly comprises of monosaccharide which are readily utilized by body and hence are advantageous from nutritional view point. Jackfruit seed contains high amount of lipid (6.74%) which is the highest in the present study. On the other hand popular delicacy water-chestnut has very low lipid content with 0.93%. Lotus seed however, contain reasonable amount of lipid (3.86%). Compared to others *N. nouchali* has much higher amount of crude fiber with 10.32% which is remarkable considering that most conventional seed and nuts contain low level of crude fiber (Table 1). Lotus seed also had reasonable amount of crude fiber (4.075%). Crude fiber is not a constituent of food in the true sense since it is not digested. Importance crude fibre in human nutrition is well known (Ladizinsky and Hymowitz 1997). In fact, a daily intake of 40g dietary fiber is recommended by Indian Council of Medical Research. In the present study except *N.nouchali* and lotus, others are poor in crude fiber. Lotus seed has been found to have a high level of total mineral in the form of ash content with 4.19% which is remarkable. By contrast conventional food grains contain much lower ash content compared to lotus seeds (Ladizinsky and Hymowitz 1997). Except *E.ferox* (0.35%) others have reasonable amount of ash content. Calorific values have been found to be high and impressive in the range of 317.78 Kcal/100gm in lotus to 376.62/100gm in Jackfruit seed with the exception of *N.nouchali* with 274.22 Kcal/100g (Table 1, Fig 1). Therefore these seeds and nuts can be considered as energy food with low fat which should be ideal for changing urban food habit. Like soluble sugar free amino

Table 1: Major nutritional components (% Dry Weight Basis) and Calorific Values of non-conventional seeds and nuts (\pm standard error of mean)

Common name	Crude protein (% dry wt.)	Total Carbohydrate (% dry wt.)	TSS (% dry wt.)	Lipid (% dry wt.)	Crude fibre (% dry wt.)	Ash content (% dry wt.)	Calorific value (Kcal/100g)
<i>T. natans</i> (Water chestnut)	10.04 ± 0.017	70 ± 0.513	6 ± 0.106	0.93 ± 0.020	1.75 ± 0.063	1.3 ± 0.041	328.56
<i>N. nouchali</i> (Mokua)	9.62 ± 0.036	51.66 ± 0.038	0.9 ± 0.020	3.23 ± 0.073	10.32 ± 0.166	1.37 ± 0.35	274.22
<i>E. ferox</i> (Makhana)	11.57 ± 0.134	70.5 ± 0.620	1.2 ± 0.017	1.9 ± 0.041	0.35 ± 0.018	0.35 ± 0.028	345.4
<i>C. argentea</i> (Chestnut)	4.4 ± 0.149	77.5 ± 0.416	1.55 ± 0.066	2.63 ± 0.064	1 ± 0.02	1.14 ± 0.026	351.32
<i>A. heterophyllus</i> (Jackfruit)	15 ± 0.089	64 ± 0.0655	5.06 ± 0.090	6.74 ± 0.120	2.6 ± 0.044	1.42 ± 0.040	376.62
<i>N. noucifera</i> (Lotus)	32.08 ± 0.018	38.66 ± 0.066	2.7 ± 0.036	3.87 ± 0.081	4.08 ± 0.071	4.2 ± 0.056	317.78
<i>G. gnemon</i>	14.78 ± 0.0444	66.87 ± 0.121	1.6 ± 0.026	1.63 ± 0.028	1.41 ± 0.032	1.1 ± 0.0330	341.3
CD at p=0.05	0.241	1.584	2.202	0.289	0.335	0.129	
CD at p=0.01	2.202	0.289	0.335	0.129	0.053	0.33	

acids can be considered as an index of nutritive value since it is readily absorbed and metabolized by body. Lotus seeds have been found to be best in the present study with 10.6 mg/gm. Among the rest *C. argentea* and *G. gnemon* also contain impressive amount of free amino acid with 8.0 and 8.2 mg/g respectively. Good amount of free amino acid is also reported for some underutilized crop seeds (Laemmli 1970). Dietary antioxidants which among others include phenolics are gaining increasing importance in recent times for their ability to scavenge free radicals (Naik and Kole 2002). They serve as exogenous non-enzymatic anti-oxidants as dietary components to reduce the risk of cancer, diabetes, coronary heart diseases and age associated oxidative stress (Prakash et al. 1988, Sherman 1952). There are reports that life style changes, mental stress, alcoholism,

nicotinism, pollution, radiation etc. also increase free radical generation in the body (Tiwary 2002). In the present study, impressive amount phenolics have been recorded for *N. nouchali* (17.6 mg/g) and *G. gnemon* (11.1 mg/g). Lotus seed also contained reasonable amount of phenolics with 7.0 mg/g. Subhasree et al. (2009) working with four leafy vegetables and Guleria et al. (2011) working with 16 herbs with medicinal values reported very impressive amount of antioxidants, phenolics and flavonoid and their efficacy were confirmed by in vitro assay. The findings of the present study are in conformity with these earlier reports and substantiate the veracity of traditional knowledge. Hence, such impressive amount of dietary antioxidants imparts nutraceutical value to these nuts and seeds.

Table 2: Protein profile of seeds and nuts of seven species resolved in 12% acrylamide gel

Species	Total proteins	Individual Protein with molecular weight (Kd)
<i>T. natans</i>	10	91.2, 74.0, 61.8, 54.7, 43.7, 41.0, 32.6, 26.5, 22.0, 19.5 No prominent band found
<i>N. nouchali</i>		No protein band could be resolved
<i>E. ferox</i>	3	95.5, 66.0, < 14.3 Most prominent band < 14.3
<i>C. argentea</i>	6	75.5, 66.0, 53.2, 41.2, 21.7 < 14.3 Most prominent bands 21.7 and < 14.3
<i>A. heterophyllus</i>	5	51.2, 41.8, 34.5, 23.0, 19.5 Most prominent bands 23.0 and 19.5
<i>N. nucifera</i>	20	> 99.0 > 99.0, 99.0, 92.5, 71.0, 66.0, 54.7, 44.3, 43.0, 41.0, 37.5 31.7, 26.5, 23.8, 22.4, 20.4, 19.0, < 14.3, < 14.3, < 14.3 Most prominent bands 92.2, 71.6 and 31.7
<i>G. gnemon</i>	4	92.2, 71.6, 61.4, 31.4 Most prominent band 31.4

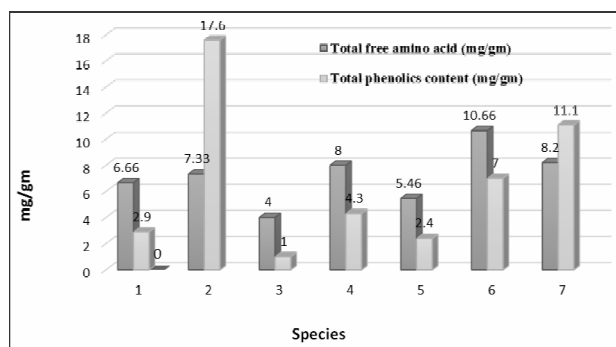


Figure: 1. Bar diagram showing Total free amino acid and Total Phenolics 1. *T. natans* 2. *N. nouchali* 3. *E. ferox* 4. *C. argentea* 5. *A. heterophyllus* 6. *N. nucifera* 7. *G. gnemon*

Seed protein profile is widely recognized as a reliable tool for plant germplasm characterization for their stability and reproducibility (Yildirim et al. 2000) and can reveal intraspecific and interspecific variation that help to resolve taxonomic and evolutionary problems. Seed protein profile was found to be highly heterogenous. Highest polymorphism was found in lotus with 20 proteins in the size range of >99.0 to <14.3 Kd with the one of 31.7 Kd being most prominent. *T. natans* also exhibited considerable polymorphism with 10 proteins in the range of 91.2 to 19.5 Kd (Table 2). The remaining four exhibited low degree of polymorphism in the range of 3 to 6 protein bands. No protein band was visible in *N. nouchali*. Probably, the extraction buffer used was ineffective. The protein profile shows that no two species have any close similarity. However all the species in the present study were unrelated phylogenetically from classical taxonomic view. Molecular analysis in terms of seed protein profile also corroborates this (Table 2).

Present study shows that contrary to general belief, non-conventional seeds and nuts are nutritionally very rich with high calorific value. Particularly lotus seeds are outstanding with 32.08% which is possibly second highest next to Soybean among seeds and seed grains. The dried powder of Lotus seed therefore can be blended with processed food to enhance their nutritive values. Lotus is famous for being India's national flower but with rich nutritive and nutraceuticals value of its seed particularly high protein content; it can be added to our food basket. Jackfruit seed, which is essentially a by-product and popular as potato substitute, is in fact superior to potato with 15.0% protein. All the seeds and nuts in the present study are low in fat content which fits well with modern urban trends and requirements for low fat consumption. Being rich in free amino acids and particularly dietary antioxidants phenolics, they possess remarkable nutraceutical value. Thus

nonconventional seeds and nuts should give priority in research and developmental activities.

सारांश

वर्तमान अध्ययन में सात असंबंधित एवं विविध प्रकार के गैर पारम्परिक बीज व गिरी जैसे— *ट्रापा नाटान्स*, *निम्फिया नौचाली*, *यूरीली फेराक्स*, *कैस्तानोप्सीस अरजेन्टिया*, *आर्द्रोकारपस हेटेरोफिलस*, *निलम्बो न्यूसीफेरा* तथा *ग्नेटम ग्नीमोन* को सम्मिलित किया गया, जिनमें पोषक मूल्य व उष्मीय मान अधिक होता है। इनमें अपक्व प्रोटीन की विविध मात्रा 4.40–32.08 प्रतिशत तथा कार्बोहाइड्रेट की विविध मात्रा 38.66–77.50 प्रतिशत पाया गया। उष्मीय मान सीमा 317.78 केसीएल/100 ग्राम से 376.62 केसीएल/100 ग्राम तथा लिपिड सीमा 0.93 से 6.74 प्रतिशत पाया गया। इनमें खाद्य आक्सी-प्रतिकारक फिनोलिक की विविधता 1.0–17.6 मिग्रा./ग्राम पाया गया। इसके अलावा इनमें मुक्त एमिनो एसिड की मात्रा सीमा 4.0–10.66 मिग्रा0/ग्राम पाया। वर्तमान अध्ययन से स्पष्ट होता है कि गैर पारंपरिक बीजों एवं गिरी में पोषक तत्वों की मात्रा अधिक होती है।

Reference

- AOAC (1975) Official Methods of Analysis 2nd Edition. Association of Official Analytical Chemists, Washington DC
- Arora RK and Pandey A (1996) Wild Edible Plants of India: Diversity, Conservation and Use. National Bureau of Plant Genetic Resources, New Delhi
- Bagchi D, Sen CK, Bagchi M and Atalay M (2004) Anti-angiogenic, antioxidant and anticarcinogenic properties of a novel anthocyanin rich berry extract formula. *Biochem* 69: 75-80
- Clegg M (1956) The application of anthrone reagent to the estimation starch in cereals. *J Food Sci Agric* 7: 40-44
- Gopalan GB, Rama Sastry V and Balasubramaniam SC (1989) Nutritive Values of Indian Foods. National Institute of Nutrition, Hyderabad.
- Guleria S, Tiku AK, Singh G and Rana S (2011) Characterization of antioxidant activity of twenty selected medicinal plants growing in North Western Himalaya. *Indian J Agric Biochem* 24(2): 117-122.
- Handique AK (2003) Nutritive values of some non-conventional leafy vegetables from ethnic sources of North East India. *Crop Res* 26(2): 361-364.
- Kaplan M, Mutlu EA, Benson M, Fields JZ, Banan A and Keshavarzian A (2007) Use of herbal preparations in the treatment of oxidant mediated inflammatory disorder. *Theor Med* 15(3): 207-216.
- Ladizinsky G and Hymowitz J (1979) Seed protein electrophoresis in taxonomy and evolutionary studies. *Theor Appl Genet* 54: 145 - 151
- Laemmli UK (1970) Cleavage of structural proteins during the assembly of head of bacteriophage T 4. *Nature* 227: 680-685.
- Naik BS and Kole C (2002) Inheritance of seed protein expression in mungbean, *Indian J Genet* 62(1): 79-80.

- Prakash D, Jain RK and Mishra PS (1988) Amino acid profile of some under-utilised seeds. *Plant Food Human Nutri* 38: 235-241.
- Sadasivam S and Manickam A (1992) *Biochemical Methods for Agricultural Science*. Wiley Eastern Ltd New Delhi
- Sherman HC (1952) *Chemistry of Food and Nutrition*. The Macmillan Company, New York.
- Singh BK, Pathak KA and Ramakrishna Y (2013) Underutilized vegetable crops and spices of Mizoram: need exploration and utilization. In: *Developing the Potential of Underutilized Horticultural Crops of Hill Regions* (Prakash N, Roy S S, Sharma P K and Ngachan S V Eds.). Today & Tomorrow's Printers and Publishers, New Delhi, pp 217-232.
- Subhasree B, Baskar R, Laxmi Keerthana R, Lijina Susan R and Rajasekaran P (2009) Evaluation of antioxidant potential in selected green leafy vegetables. *Food Chem* 115: 1213-1220.
- Tiwary AK (2001) Imbalance in antioxidant defense and human disease: Multiple approach of natural antioxidant therapy. *Curr Sci* 81: 1179-1187.
- Wang ZY (2005) Impact of anthocyanin from *Malva sylvestris* on plasma lipid and free radical. *J Forest Res* 16: 228-232.
- Yildirim A, Mavi A, Oktay M, Kara AA, Algur OF and Bilalogu V (2000) Comparison of antioxidant and antimicrobial activities of tilia (*Tilia argentea* Desf Ex DC), sage (*Salvia triloba* L.) and black tea (*Camelia sinensis*) extracts. *J Agric Food Chem* 48: 5030-5034.