Phytochemical Qualitative Analysis and Total Tannin Content in the Aqueous Extract of Areca catechu Nut



INTRODUCTION:

Rural areas of many developing countries rely on the herbs for medicines. People living in rural areas know the value of these herbal medicines when used in therapeutic doses but they do not actually understand the science behind them [1 and 2]. Plant constituents are isolated either directly or indirectly and used for the synthesis of various drugs. *Areca catechu* is commonly called as betel-nut belonging to the family Arecaceae cultivated in tropical Pacific, Asia and parts of East Africa. *Areca catechu* is chewed with betel leaf and lime for health and cultural aspects. *Areca catechu* is commercially available in the market as dried, cured and fresh forms. *Areca* nut when chewed with betel leaf acts as a mild stimulant [3].

Many phytochemicals present in the plants has long term beneficial effects when consumed by humans. The phytochemicals appear to have structure and mode of action distinct from those of the antibiotics in current use suggesting the cross resistance with agents already in use. Hence, it is significant to study the phytochemicals and their activity. The phytochemicals prevent diseases in man and they act as prophylactic and therapeutic agents in cardio protection [4]. Constituents of *Areca catechu* are known for its beneficial effect on skin, suggesting its use in cosmetic industries [5-8].

The pharmacological effect of Areca nut was investigated

by the several researchers [9-11]. *Areca catechu* seed is also used in folklore medicine for the treatment of various ailments. The chemical constituents of the nut are carbohydrates, fats, proteins, polyphenols, crude fibre, alkaloid and minerals. Flavonols and tannins constitute the large proportion in dry *Areca*. Polyphenols are the responsible constituent for the astringent taste of *Areca* [12]. Hence in the present investigation, various phytochemicals and total tannin content are analysed in the aqueous extract of *Areca catechu* nut.

MATERIALS AND METHODS

Collection and authentication of plant material:

Commercially sold *Areca catechu* nut in the form of flakes known as 'Seeval' in Tamil was purchased in large quantity from the local market and was authenticated by Dr. R. Pandian, Head of the Department of Plant Biology and Biotechnology, Presidency College, Chennai – 600 005.

Preparation of Areca catechu nut powder:

The market sample of *Areca catechu* nut was ground into coarse powder. Then, it was stored in an air tight container for further extraction procedures.

Preparation of Areca catechu nut aqueous extract:

The *Areca catechu* nut powder (100 grams) was soaked in 2 liters of double distilled water for 24 hours at room temperature. Then, the content was filtered and concentrated

Vanimakhal R. R

Post Graduate and Research, Department of Zoology, Ethiraj College for Women, Chennai-8, Tamil Nadu, India. Email: vani.reva@gmail.com; Phone No: +91 9789929079



Conflict of interest: Authors reported none

^{*}Corresponding author:

in a water bath at 100 $^{\circ}$ C until a semi solid residue was obtained. The weight of the extract was noted and the yield was stored in the refrigerator.

Phytochemical analysis in the aqueous extract of *Areca catechu* nut:

Qualitative phytochemical screening of secondary metabolites was performed using standard protocol [13 and 14]. Quantitative estimation of total tannin in the aqueous extract of *Areca catechu* nut extracts:

The quantitative analysis of total tannin content in the aqueous extract of *Areca catechu* nut was determined by Folin-Denis method [15] with minor modifications. A stock solution of *Areca catechu* nut aqueous extract was prepared in the concentration of 1 mg/ml. For quantification, to 0.1 ml of *Areca catechu* nut aqueous extract, 1 ml of distilled water and 0.5ml of Folin-Denis reagent were added and mixed thoroughly. This mixture was alkalinized by adding 1 ml of 15% (w/v) sodium carbonate and kept in dark for 30 minutes at room temperature. The absorbance was read at 700 nm. Pure tannic acid (1 mg/ml) was used as standard. Blank consists of all reagents except the extract. Result was expressed as mg tannic acid equivalent per gram of dry weight (mg TE/g) of extract. The total tannin was calculated by using the formula

Total tannin content = TE x V/m

Where, TE = Tannic acid equivalence (mg/ml), V =Volume of the extract (ml)

m = Weight of the pure plant extract (g)

RESULTS

Phytochemical screening of secondary metabolites revealed the presence of saponin, flavonoid, tannin, phenol, steroid, alkaloid and protein. Whereas, acid and triterpenoid recorded their absence in the aqueous extract of *Areca catechu* nut (Table 1).

As tannin is the second main constituent of *Areca catechu* nut after alkaloids it was proposed to quantify the total tannin content in the aqueous extract of *Areca catechu* nut and the total tannin content was found to be 95.3 mg TE/g.

Table 1: Qualitative analysis of secondary metabolites in the aqueous	
extract of Areca catechu nut	

S.No	Test name	Secondary metabolites	Aqueous extract
1.	Foam test	Saponin	+
2.	Sodium bicarbonate test	Acid	-
3.	Shinoda test	Flavonoid	+
4.	Lead acetate test	Tannin	+
5.	Ferric chloride test	Phenol	+
6.	Lieber- mann-Burchard test	Steroid	+
7.	Hager's test	Alkaloid	+
8.	Salkowski test	Triterpenoid	-
9.	Biuret test	Protein	+

(+) Present; (-) Absent

DISCUSSION

Arecaceae family is also called as palmaceae in historical days. The pharmacological effects of Arecaceae have been reported earlier [16]. Phytochemical screening from various medicinal plant extracts revealed their medicinal and physiological values [17]. There are more than thousand known phytochemicals that have protective or disease preventive properties. These phytochemicals are present in the plants to protect them against diseases. Recent researches demonstrate that these phytochemical can also protect human from various diseases when consumed. Most of the phytochemicals are present in greens, vegetables, fruits and nuts. Tannin compounds play a major role in plants to protect them from predators and also in growth regulation [18]. In human, it promotes free radical scavenging activity and inhibits the tissue damage when compared to vitamin C and E [19]. The optimal dose of tannin promotes better health in human. In Areca, the tannins are reported to be present in the condensed form [20-22]. Tannin possess scavenging activity on free radicals, hence it serves as a good antioxidant. The present investigation revealed the medicinal properties of Areca catechu nut.

CONCLUSION

Many herbal plants are tested for their medicinal properties and used for the pharmaceutical preparations to treat human beings against several diseases. Phytochemicals are natural bioactive compounds present in various plant parts which help in treating numerous diseases. *Areca catechu* nut found to possess various phytochemicals revealing the therapeutic value.

REFERENCES

- Maheshwari SC, Gill R, Maheshwari N, Gharyal PK. The isolation and culture of prtotoplasts; in differentiation of protoplasts and transformed plant cells (eds.) J Reinert and Binding (New York: Springer-Verlag) 1986; 20–48.
- 2. Van Wyk BE, Gericke N. People's Plants. A guide to useful plants of Southern Africa. Pretoria: Briza Publications; 2000.
- Gupta PC, Ray CS. Epidemiology of betel quid usage Review Article. Ann Acad of Med. 2004; 33(4): 31S–36S. http://www.ncbi.nlm.nih.gov/pubmed/15389304
- Vasanthi HR, Shrishrimal N, Das DK. Phytochemicals from plants to combat cardiovascular disease. Curr Med Chem. 2012; 19(14): 2242–2251. http://www.ncbi.nlm.nih.gov/ pubmed/22414106
- Padmaja PN, Bairy KL, Kulkarni DR. Pro-healing effect of betel nut and its polyphenols. Fitoterapia, 1994; 65: 298–300.
- Lee KK, Choi JD. The effects of Areca catechu L. extract on anti-inflammation and anti-melanogenesis. Int J Cosmetic Sci. 1999; 21: 275–284. http://www.ncbi.nlm.nih.gov/ pubmed/18503445
- 7. Lee KK, Cho JJ, Park EJ, Choi JD. Anti-elastase and anti-hyaluronidase of phenolic substance from *Areca catechu* as a new anti-ageing agent. Int J Cosmetic Sci. 2001; 23: 341–346. http://www.ncbi.nlm.nih.gov/pubmed/18498484
- Ashawat MS, Shailendra S, Swarnlata S. *In vitro* antioxidant activity of ethanolic extracts of *Centella asiatica*, *Punica granatum*, *Glycyrrhiza glabra* and *Areca catechu*. Res J Med Plant, 2007; 1: 13–16.
- Pithayanukul P, Nithitanakool S, Bavovada R. Hepatoprotective potential of extracts from seeds of *Areca catechu* and nutgalls of *Quercus infectoria*. Molecules, 2009; 14(12): 4987– 5000. http://dx.doi.org/10.3390/molecules14124987
- 10. Shrestha J, Shanbhag T, Shenoy S. Antiovulatory and abortifacient effects of *Areca catechu* (betel nut) in female rats.

Indian J Pharmacol. 2010; 42: 306-311._ http://dx.doi. org/10.4103/0253-7613.70350

- Khan S, Mehmood MH, Ali ANA, Ahmed FS, Dar A, Gilani AH. Studies on anti-inflammatory and analgesic activities of betel nut in rodents. J of Ethnopharm. 2011; 135: 654–661. http://dx.doi.org/10.1016/j.jep.2011.03.064
- Raghavan V, Baruah H. Areca nut: India's popular masticatory-History, chemistry and utilization. Econ Bot. 1958; 12: 315–345.
- Overtone H. Isolation, purification and preliminary observation in elucidation of structures by physical and chemical methods. Bentley, K.W. (Ed.), Interscience Pub. New York: 1963; 34.
- 14. Harborne JB. Phytochemicals Methods. Chapman and Hall Ltd. London. 1973; 49–188.
- Schanderi SH. Methods in food analysis. Academic Press, New York, USA: 1970.
- Patil PR, Rakesh SU, Dhabale PN, Burade KB. Pharmacological activities of *Areca catechu* Linn. -Review Article. J Pharm Res. 2009; 2: 683–687.
- 17. Sofowora A. Medicinal plants and traditional medicine in Africa. Ibadan, Nigeria: Spectrum Books Ltd. 1993; 191–289.
- Katie EF, Thorington RW. Squirrels: The animal answer guide. Baltimore: Johns Hopkins University Press. 2006; 91.
- 19. Bagchi K, Puri S. Free radicals and antioxidants in health and disease. East Mediterranean Health J.1998; 4: 350–360.
- Govindarajan VS, Mathew AG. Polyphenolic substances of areca nut. Phytochemistry, 1963; 2: 321–326.
- Majumder AM, Kapadi AH, Pendse GS. Chemistry and pharmacology of betel nut *Areca catechu* Linn. J Plant Crops, 1979; 7: 69–92.
- 22. Wang CK, Lee WH. The separation, characteristics and biological activities of phenolics in areca fruit. J Agric Food Chem. 1996; 44: 2014–2019.