



IJPPR

INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals

ISSN 2349-7203



Human Journals

Research Article

October 2015 Vol.:4, Issue:3

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DPPH Free Radical Scavenging Activity of *Myristica fragrans* Houtt. Fruit

 **IJPPR**
INTERNATIONAL JOURNAL OF PHARMACY & PHARMACEUTICAL RESEARCH
An official Publication of Human Journals

ISSN 2349-7203

 HUMAN

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Submission: 29 September 2015
Accepted: 3 October 2015
Published: 25 October 2015

Keywords: *Myristica fragrans* Houtt., antioxidant activity, ascorbic acid, DPPH, methanolic extract

ABSTRACT

Myristica fragrans Houtt. of family Myristicaceae is commonly known as nutmeg (Jaiphal in Gujarati). Various parts of tree has been used in traditional folkloric medicine. The present study was concentrated on the *in-vitro* antioxidant methods like DPPH free radical assays. The methanolic extract of *Myristica fragrans* Houtt. fruit was subjected to the DPPH method. The results of antioxidant activity revealed that, the methanolic extract shows good IC₅₀ values. The results were compared with the standard ascorbic acid. The plant contains flavonoids, alkaloids, saponins and tannins. These active constituents alone or in combination may be responsible for the observed antioxidant activity.



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INTRODUCTION

Natural antioxidants, particularly in fruits and vegetables have gained increasing interest among consumers and the scientific community because epidemiological studies have indicated that frequent consumption of natural antioxidants is associated with a lower risk of cardiovascular disease and cancer (Renaud *et al.*, 1998; Temple, 2000).

Herbal plants are known to contain a variety of antioxidants. Numerous substances have been suggested to serve as antioxidants. It has been revealed that various phenolic antioxidants, such as flavonoids, tannins, coumarins, xanthenes and more recently procyanidins scavenge radicals dose-dependently, thus they are viewed as promising therapeutic drugs for free radical pathologies. (Halliwell *et al.*, 1998; Azam *et al.*, 2004).

Myristica fragrans Houtt. is an evergreen tree, native of the Moluccas (or Spice Islands) of Indonesia and cultivated throughout Malaya. The seed of the plant is known as “nutmeg” and the arillus of the seed is called “mace”. Both nutmeg and mace contain many volatile oils. These oil constituents have a variety of individual pharmacological effects, some of which oppose others (Jellin *et al.*; 2005). The fruit contains ethereal oil-cells often with phenolic and myristicin; the seed and the aril are used for flavouring food. Of the 72 species of *Myristica fragrans* of global level distribution, five species are reported in South India (Gamble, 1921). Nutmeg also contains fat or butter that has applications in pharmaceutical preparations. Sabinene, myristican, safrole and elemicin constitute 80% of both these oils. The West Indian oils (oils from nutmeg cultivated in Grenada) have considerable amounts of α -pinene, β -pinene and sbinene (40%-50%) and are low in safrole and myristicin, whereas East Indian oils (oils from nutmeg cultivated in Indonesia and other regions in South East Asia) have higher amounts of myritican (Purseglove *et al.*, 1981; Maya *et al.*, 2004). Nutmeg, mace and their oils are mostly used with fresh foods like cakes, biscuits, doughnuts, fruit pies, egg nog and puddings to give them a delicate smooth flavor. The oil is used in canned soups and stews, and has an important application in neutralizing the unpleasant odor of cooked Cabbage. The present work was undertaken to evaluate nutmeg germplasm for chemical composition and to shortlists accessions suitable for the nutrient industry.

MATERIALS AND METHODS

Plant material

Myristica fragrans Houtt. was obtained from local market store Shaibaug Ahmedabad Gujarat.

Preparation of extract

The fruits were washed with distilled water, oven dried and well powdered in mixture. 10 g fruit powdered material successively extracted with 100 ml methanol overnight by Rotary Vacuum Evaporator. The resultant extracts were filtered and evaporated to dryness at 35⁰C in *vacuo*, and used for the antioxidant activity.

Antioxidant potentialities

2, 2- Diphenyl-1- picrylhydrazyl (DPPH) and ascorbic acid were obtained from Hi media, India. DPPH method used (Fogliano *et al.*, 1999) was adopted with suitable modifications to our particular circumstances. Methanolic solution of DPPH (2 mg/ 50 ml) was used.

DPPH free radical scavenging activity

Antioxidant activity measured by using DPPH free radical scavenging assay method (Fogliano *et al.*, 1999). Percent inhibition of DPPH was calculated by the following equation (Lee *et al.*, 1998):

$$\% \text{ Inhibition} = 1 - (A1/A2) \times 100$$

Where, A1 is the absorbance of the test sample, A2 is the absorbance of control reaction. After that IC 50 value was calculated.

RESULTS AND DISCUSSION

Myristica fragrans Houtt. methanolic extract was reddish brown in color. Antioxidant activities by DPPH assay of *Myristica fragrans* Houtt. extracts are given in Table no 1. In *Myristica fragrans* Houtt. methanolic extract showed highest antioxidant activity (94.295%) at 0.25 mg/ml. When it compares to the standard ascorbic acid it shows 3.4% less antioxidant activity

at 0.25 mg/ml concentration. But it shows higher antioxidant activity compared to the standard Ascorbic acid at 0.001 mg/ml concentration. It means lower concentration shows higher antioxidant activity compared to the standard Ascorbic acid.

Table no 1. Antioxidant activity of *Myristica fragrans* Houtt. by DPPH assay

<i>Myristica fragrans</i> Houtt.			Ascorbic acid
Sr.No	Concentration (mg/ml)	% inhibition	% inhibition
1	0.25	94.295±0.085	97.78±00.00
2	0.125	88.17±0.17	97.70±0.00
3	0.062	75.44±0.13	97.19±0.00
4	0.031	66.93±0.04	92.17±0.00
5	0.015	61.655±0.125	77.44±0.00
6	0.007	53.525±0.255	47.65±0.00
7	0.003	48.765±0.085	33.87±0.00
8	0.001	43.995±0.085	27.57±0.00

DPPH is a common and recently used method. This method is simple, rapid and sensitive for antioxidant activity. In addition ascorbic acid was also measured to compare antioxidant activity of methanol extract of *Myristica fragrans* Houtt. DPPH measurement expressed as IC 50 i.e. amount of extract concentration (mg/ml). Which scavenge DPPH radicals by 50%. IC 50 value of methanol extract of *Myristica fragrans* Houtt. is 0.0065 and methanolic extract of ascorbic acid is 0.0105. *Myristica fragrans* Houtt. extract has the highest scavenging ability to 50% DPPH radical, which requires extract concentration 0.0065 mg/ml. High antioxidant capacity of fruit extract is due to the presence of tannin, flavonoids, and terpenoids compounds. These compounds contribute to serve as electron donors as described by (Buhler and Miranda, 2000; Joshi *et al.*, 2008 and Das *et al.*, 2011).

CONCLUSION

The hazardous effects of synthetic antioxidants and the emergence of antibiotic resistant strains have reviewed the search for antioxidant agents from natural sources. From different studies

conducted it has been found that fruits hold a tremendous potential to serve as a source of newer, effective, safer and better antioxidant agents. The tested extracts exhibited strong scavenging activity against DPPH radicals. From the study it is concluded that the antioxidant capacity of *Myristica fragrans* Houtt., which are widely used all over the world are considered as good sources of antioxidants as observed in DPPH scavenging assay. It also proves its use as food in the daily diet of people with nutritional and therapeutic value, can be used as an accessible source of natural antioxidants with consequent health benefits particularly. *Myristica fragrans* Houtt. has their higher antioxidant activity relatively standard ascorbic acid also have their higher antioxidant activity that can be consider as a model herbal drug for experimental studies including free radical induced disorders like cancers, diabetes, aging, and cardiovascular diseases.

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