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ARTICLE

Chemistry, antioxidant and antimicrobial potential of nutmeg (*Myristica fragrans* Houtt)

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KEYWORDS

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Abstract Antioxidant and antimicrobial activities of nutmeg (*Myristica fragrans* Houtt) seed extracts were evaluated. Seeds were extracted with acetone, ethanol, methanol, butanol and water. All the extracts have shown significant antioxidant and antimicrobial activities against the tested microorganisms. Among all extracts, acetone extract has shown the highest antioxidant activity. The acetone extract showed 93.12 ± 1.48 mg gallic acid equivalents (GAE)/100 g dry weight total phenolic content, DPPH scavenging activity of 63.04 ± 1.56%, chelating activity of 64.11 ± 2.21% and 74.36 ± 1.94% inhibition of β-carotene bleaching, at 1 mg/mL extract concentration. Out of all extracts, acetone extract was able to exert antimicrobial activity against all tested bacteria and fungi. Acetone extract has shown the strongest antibacterial and antifungal activity with *Staphylococcus aureus* (13.8 ± 0.42 mm) and *Aspergillus niger* (14.4 ± 0.37 mm), respectively. GC–MS analysis of acetone extract has revealed the presence of 32 compounds of extract representing 99.49%. Sabinene (28.61%) has shown the highest occurrence in the extract. β-Pinene (10.26), α-pinene (9.72), myristicin (4.30%), isoeugenol (2.72%), p-cymene (1.81%), carvacrol (1.54%), eugenol (0.89%) and β-caryophellene (0.82%) were reported as possible contributor for antioxidant and antimicrobial activity of nutmeg.

1. Introduction

Free radicals and other reactive oxygen species like hydroxyl radical (·OH), superoxide anion ($\cdot O_2-$), singlet oxygen ($^1O_2$) and hydrogen peroxide (H_2O_2) can cause oxidative damages to biological macromolecules which can lead to initiation and/or progression of various diseases [13] such as cellular and metabolic injury, cancer, atherosclerosis, inflammation,

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aging, immunosuppresion, diabetes, ischemic heart disease and neurodegenerative disorder such as Alzheimer's and Parkinson's disease [1]. Exogenous antioxidants may contain various compounds such as vitamin C, tocopherols, carotenoids, flavonoids and variety of phenolic compounds which could prevent human beings from various diseases resulting due to oxidative stress. Most widely used synthetic antioxidant compounds were butylated hydroxytoluene (BHT) and butylated hydroxyanisole (BHA) which have been restricted from usage because of their liver damage and carcinogenic potential. Therefore, in present scenario strong emphasis has been given to search for new and natural antioxidants from dietary plants because they can safeguard human body from various diseases caused by oxidative damages of lipid, protein and nucleic acid.

Furthermore we know that living organisms including humans are constantly exposed to potential harmful pathogens throughout their life which results in various diseases and have great impact on human health. The human beings have evolved defense mechanism to overcome these problems but often, this first line of defense mechanism get failed and various chemotherapeutic agents are used to combat the infections caused by pathogens. In past, a large number of chemical agents have been discovered or synthesized in order to treat and cure these infections but widespread and indiscriminate use of these drugs has led to the development of many drug resistant strains which constitutes a major problem worldwide as the existing drugs are becoming ineffective to control them. Consequently, there is an urgent need to look for alternatives of synthetic antibiotics and other drugs.

The possible solution for the aforesaid problems could come from spices and aromatic herbs which have been reported to have antioxidant and antimicrobial properties [15,37] besides their numerous folk medicinal usages. Nutmeg is dried kernel of broadly ovoid seed of *Myristica fragrans* Houtt (Family: Myristicaceae). Nutmeg is widely used as spices and in alternative medicine as it has been reported to have aphrodisiac [31], memory enhancer, antidiarrheal property [14], anti inflammatory and anti-cancer property [23]. The main reason to choose nutmeg for this study is that, in spite of traditional use in numerous medical conditions, nutmeg has not been comprehensively evaluated for their antioxidant and antimicrobial potential, which could be contributed by the variety of active phytochemicals including vitamins, carotenoids, terpenoids, alkaloids, flavonoids, lignans and phenolics, etc. These compounds render their effects via different mechanisms such as radical scavenging, metal chelation, inhibition of lipid peroxidation and quenching of singlet oxygen to act as antioxidants.

So far researchers have analyzed essential oil, lignans and volatile aglycones from nutmeg and mace for its composition, antioxidant properties and antimicrobial activity [16,5]. The present study is an important report for antioxidant and antimicrobial activity of nutmeg using various extracts i.e. acetone, ethanolic, methanolic, butanolic and aqueous extracts of nutmeg. The selection of the various solvents is based on the difference in their polarities. The main aim to choose different solvents is that maximum solubility of different constituents of nutmeg powder will depend upon polarity of solvents. The main objectives of present study were (i) to evaluate antioxidant potential of various extracts of nutmeg (ii) to investigate antimicrobial activity of various extracts of nutmeg against pathogenic bacteria and fungi, (iii) to evaluate the minimum inhibitory concentration (MIC), (iv) identification of compounds responsible for strong antioxidant and antimicrobial activity by GC–MS.

2. Materials and methods

2.1. Sample preparation

The fully matured seeds of nutmeg were procured from R.S. Spices Store, Moradabad, Uttar Pradesh and specimens are preserved in the institute herbarium. Thoroughly washed nutmegs were dried in hot air oven at 40 °C for 72 h and grounded to a fine powder which was kept in an air-tight container at 4 °C until further use. For the aqueous extraction, 10 g of fine powder was extracted with 100 mL distilled water at 90 °C for 30 min in water bath. Ten grams of powdered sample was put into a 500 mL bottle and 100 mL of acetone or butanol or ethanol or methanol was added to each bottle. After 72 h of storage at room temperature, the supernatant and the sediment were separated by vacuum-filtration. The supernatant was dried by vacuum-evaporator and stored at −20 °C until further used.

2.2. Determination of total phenolics

Phenolic contents of the various extracts of nutmeg were determined according to the Folin–Ciocalteu colorimetric method [27]. Gallic acid was used for calibration of standard curve. The results were expressed as mg gallic acid equivalents (GAE)/100 g dry weight of plant material.

2.3. Measurement of reducing power

The reducing power of various extracts (0.025–2 mg/mL) or BHT (0.025–1.0 mg/mL) was determined according to the method given by Yen and Chen [38]. BHT was used as positive reference compound.

2.4. DPPH radical scavenging activity

The DPPH (1,1-diphenyl-2-picrylhydrazyl) radical scavenging activity of the various extract (0.025–2 mg/mL) or BHT (0.025–1.0 mg/mL) was measured using the method of Brand-Williams, Cuvelier, and Berset [3]. BHT was used as positive control.

2.5. Chelating activity on Fe^{2+}

The chelating ability of ferrous ions by extracts of nutmeg or EDTA (0.20–1.2 mg/mL) was estimated by the method given by Dinis, Madeira, and Almeida [10]. EDTA served as the positive control.

2.6. Determination of β-carotene bleaching activity

Antioxidant activity of extracts of nutmeg (1 mg/mL) was evaluated using β-carotene-linoleate model system, as described by Sun and Ho [30]. BHT was used as positive control.

2.7. Antimicrobial activity

In the present study *Bacillus subtilis* (MTCC 441), *Staphylococcus aureus* (MTCC 737), *Pseudomonas putida* (MTCC

1072) and *Pseudomonas aeruginosa* (MTCC 7903) were used as test bacteria and *Aspergillus fumigatus* (MTCC 343), *Aspergillus niger* (MTCC 282), *Aspergillus flavus* (MTCC 277) were used as test fungi. All microbial cultures were obtained from Institute of Microbial Technology, Chandigarh, India. Nutrient agar (NA) and potato dextrose agar (PDA) was used for culturing of test bacteria and fungi, respectively. All strains were stored at −20 °C in the appropriate medium containing 10% glycerol and regenerated twice before use in the experiments.

Disk diffusion method was used to determine antimicrobial activity of various extracts of nutmeg using 5 μL of 10 mg/mL extracts dissolved in dimethylsulfoxide (DMSO). DMSO was used as negative control [28]. Gentamicin (30 μg/disc) and nystatin (30 μg/disc) were used as positive reference for bacteria and fungi, respectively. Antimicrobial activity was evaluated by measuring zone of inhibition against the test organisms. MIC of the various extracts of nutmeg was determined by micro-well dilution method against test bacterial and fungal strains [28].

2.8. Analysis of acetone extract by Gas Chromatography–Mass Spectrometry (GC–MS)

GC–MS analysis of extract showing most promising antioxidant and antimicrobial activity i.e. acetone extract was carried out using Perkin Elmer Clarus-500 gas chromatograph mass spectrometer provided with a FID detector and 'Elite-5' capillary column [(5% diphenyl)-dimethylpolysiloxane], length 30 m, id., 0.25 mm and film thickness 0.25 μm. Ultra pure nitrogen was used as carrier gas (flow rate, 1 mL/min). Injector and interface temperature were 200 and 210 °C, respectively. Column temperature was programmed from 60–280 °C at an increasing rate of 10 °C/min, held at initial and final temperature for 2 min. The major peaks were analyzed by comparing its mass fragments patterns with the standard spectra available in Perkin Elmer GC–MS NIST library.

3. Results and discussion

3.1. Total phenolic content

Total phenolic content of nutmeg extracts was determined according to the Folin–Ciocalteu method and expressed as mg GAE/100 g dry weight of plant material. Total phenolic content was found to be affected by the solvents used for extraction. Acetone was found to be significantly the most effective solvent for extraction of phenolic compounds than the other solvents used in the study ($p < 0.05$). The trend of phenolic content was reported to be acetone > ethanol > methanol > aqueous > butanol. Total phenolic content of acetone extract was found to be 93.12 ± 1.48 while least phenolic content i.e. 49.82 ± 1.26 mg GAE/100 g dry weight was reported from butanol extract (Table 1). It has been reported that total phenolic content and antioxidant activity have significant and positive correlation [35]. In plants, phenolic antioxidants are produced primarily by secondary metabolism and their antioxidant properties mainly depends on redox properties and chemical structure i.e. number and position of hydroxyl group which play important role in scavenging free radical, chelating transitional metals and inhibiting lipoxygenase [9]. It has been reported that upon excessive drying, antioxidant activity decreases significantly which could be attributed to the interaction of oxidized phenolics or free phenols with cell polysaccharides upon drying which is reinforcing the significance of using fresh spices [7].

3.2. Reducing power of various extracts of nutmeg

The reducing power of a compound may serve as a significant indicator of its potential antioxidant activity. The presence of reductants in the extracts of nutmeg causes the reduction of Fe^{3+}/ferricyanide complex to ferrous form by donating an electron. The amount of Fe^{2+} complex can be monitored by measuring the formation of perl's prussian blue at 700 nm wavelength. Increase in absorbance at 700 nm reflects an increase in reducing ability. Acetone extract has shown the highest reducing power but was significantly lower than positive standard BHT ($p < 0.05$). At extract concentration, 1.0 mg/mL, highest absorbance at 700 nm was reported from acetone extract (0.75 ± 0.013), least absorbance was reported from butanol extract (0.33 ± 0.019) whereas absorbance for positive standard (BHT) was reported to be 1.41 ± 0.032, at same concentration (Fig. 1). At this concentration, the absorbance of BHT was 1.88-fold higher to acetone extract. The reducing power of various extracts of nutmeg was found to be concentration dependent (for acetone extract: $R^2 = 0.9993$). Trend of reducing power was reported to be BHT > acetone > ethanol > methanol > aqueous > butanol. EC_{50} value was found to be 0.36 ± 0.013 and 0.65 ± 0.024 mg/mL for BHT and acetone extract, respectively. In general, extracts with high total phenolic content presented lower EC_{50} values in reducing power assay (Table 1).

3.3. DPPH radical scavenging activity

Antioxidant activity of the various extracts of nutmeg was tested by the DPPH radical scavenging assay where the consumption of a stable free radical (DPPH·) was measured. When a solution of DPPH is mixed with an antioxidant compound which can donate a hydrogen atom, it gives rise to the reduced form, diphenypicrylhydrazine (non radical). The color of reaction mixture changes from purple to yellow and its absorbance at wavelength 517 nm decreases. Antioxidant activity of various extracts was compared with a known high antioxidant activity standard compound i.e. BHT. Fig. 2 shows the dose response curves of the various extracts of nutmeg. At 1 mg/mL concentration, highest free radical scavenging activity was observed from acetone extract (63.04 ± 1.29%), least activity was observed from butanol extract (36.21 ± 1.31%) where as free radical scavenging activity of BHT (positive control) was found to be 94.2 ± 1.41% ($P < 0.05$). It was found that radical scavenging activity of the extracts increased with increasing concentration (for acetone extract: $R^2 = 0.8924$). Lower IC_{50} value indicates higher antioxidant activity. IC_{50} value for BHT was reported to be 0.17 ± 0.018 mg/mL whereas for acetone extract it was found to be 0.66 ± 0.015 mg/mL (Table 1). It has been reported that after absorption nutmeg lignans and their glycosides are metabolized to produce biologically active compound containing catechol structure which could account for the high antioxidant potential of the nutmeg [19]. It has been reported that compounds having catechol structure are considered as good antioxidants because catechol structure can easily donate

Table 1 Total phenolic content, EC_{50} values (mg/mL) and IC_{50} vaules (mg/mL) of different extracts of nutmeg.[a]

Sample	Total phenol content (mg GAE/100 g)	Reducing power (EC_{50})	DPPH Scavenging activity (IC_{50})
BHT	Ns[b]	0.36 ± 0.013	0.17 ± 0.018
Acetone	93.12 ± 1.48	0.65 ± 0.024	0.66 ± 0.015
Ethanol	70.69 ± 2.06	0.78 ± 0.041	0.77 ± 0.011
Methanol	61.26 ± 1.32	0.91 ± 0.017	1.04 ± 0.029
Aqueous	57.49 ± 1.87	1.09 ± 0.014	1.18 ± 0.022
Butanol	49.82 ± 1.26	1.46 ± 0.021	1.40 ± 0.018

[a] Values represent mean ± SD ($n = 3$).
[b] Not studied.

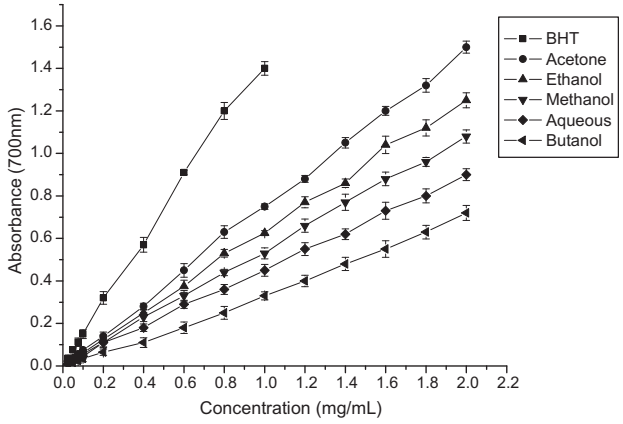

Figure 1 Reducing power of various extracts of nutmeg. BHT was used as positive reference. Vertical bars represents SD ($n = 3$).

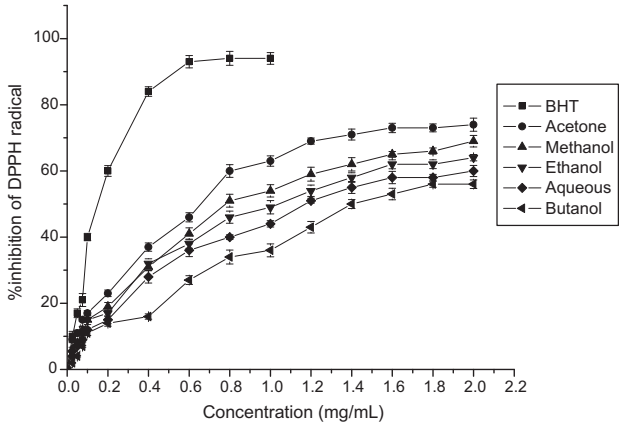

Figure 2 Inhibition of DPPH radical by various extracts of nutmeg. BHT was used as positive reference. Vertical bars represents SD ($n = 3$).

phenolic hydrogen or electron to the acceptors like reactive oxygen species or lipid peroxyl groups [26].

3.4. Chelation activity on Fe^{2+}

Iron is an important transition metal and it is required for normal cell physiology but Fe^{2+} can accelerate lipid oxidation because it can break lipid peroxide and hydrogen peroxide into reactive free radicals via Fenton reaction ($Fe^{2+} + H_2O_2 \rightarrow Fe^{3+} + {}^{\cdot}OH + OH^-$). Highly reactive hydroxyl radical can cause oxidative stress which eventually leads to damage of cellular lipids, protein and nucleic acid. In this study we have tested the chelation of Fe^{2+} by extracts of nutmeg because chelation will remove free iron from circulation and could present a promising approach to curtail oxidative stress induced diseases. At 1.0 mg/mL concentration, highest chelating activity was shown by ethanol extract (72.11 ± 1.54%) whereas least cheating activity was shown by aqueous extract (48.34 ± 1.85%) while positive control, EDTA has shown 99.6 ± 1.65% chelation activity at the same concentration ($P < 0.05$, Fig. 3). All the extracts demonstrated an ability to chelate iron (II) ions in a dose-dependent manner (for ethanol extract: $R^2 = 0.9569$). Common trend for chelating activity was observed as EDTA > ethanol > acetone > methanol > butanol > aqueous. It has been observed that chelating agents which form σ bond with metal can work as secondary antioxidants because they reduce redox potential and stabilize oxidized form of metal ion [29].

3.5. β-Carotene bleaching assay

In β-carotene bleaching assay, oxidation of linoleic acid produces hydroperoxides as free radicals upon incubation at 50 °C. These hydroperoxides attack highly unsaturated β-carotene molecule, oxidize them and as a result, β-carotene molecules undergoes rapid decolorization. The corresponding decrease in absorbance can be monitored spectrophotometrically. The presence of antioxidants in the extract can reduce the extent of β-carotene bleaching by acting on the free radi-

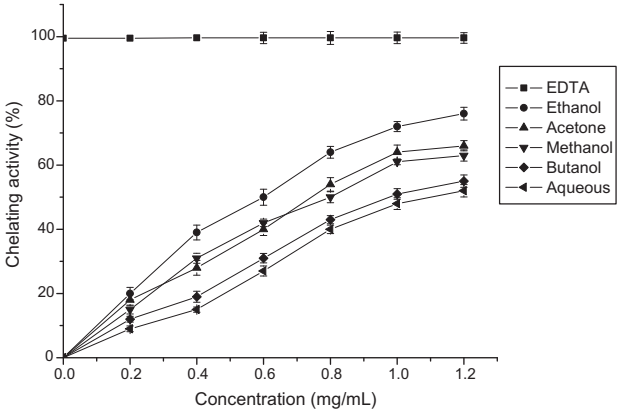

Figure 3 Percentage chelating activity of various extracts of nutmeg. EDTA was used as positive reference. Vertical bars represents SD ($n = 3$).

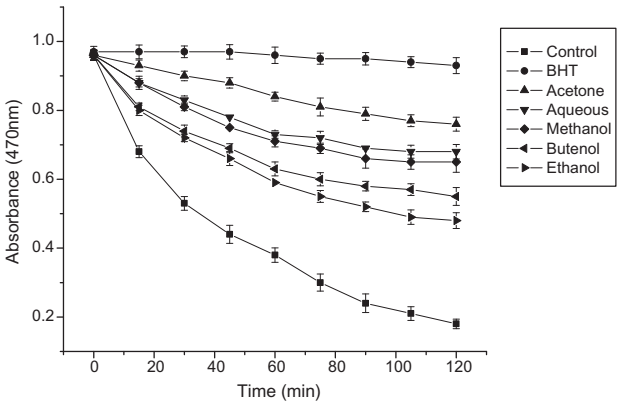

Figure 4 Antioxidant activity of various extracts (1.0 mg/mL) of nutmeg measured by β-carotene bleaching method. BHT was used as positive reference. Vertical bars represents SD (n = 3).

cals formed in the system. Oxidation rate of β-carotene holds an inverse relationship with antioxidation activity of the extract. The antioxidant activity of different extracts of nutmeg was assayed in the β-carotene-linoleate model system and compared with BHT (Fig. 4). The addition of 1.0 mg of extract of nutmeg or BHT was showing good effect on inhibition of oxidation of linoleic acid and subsequent bleaching of β-carotene, in comparison to the control which contained no antioxidant component ($P < 0.05$). All the extracts were less effective in comparison to BHT (94.9 ± 1.03%), but showed good inhibitory activity. Among extracts, acetone extract has shown the highest activity (74.36 ± 1.94%) while the least activity was observed in ethanol extract (38.47 ± 1.31%). The common trend in inhibition of β-carotene bleaching was observed as BHT (94.91 ± 1.03%) > acetone (74.36 ± 1.94%) > aqueous (64.11 ± 1.43%) > methanol (60.26 ± 1.08%) > butanol (47.44 ± 1.27%) > ethanol (38.47 ± 1.31%). These results indicate that all the extracts have acted as an effective antioxidant in β-carotene linoleic acid model system. In this assay, high antioxidant activity could be attributed to isoeugenol, lignans, eugenol and β-caryophyllene which have been isolated from nutmeg by researchers [16,5,12]. Isoeugenol and lignans are important because being less polar they will have high partition coefficient and can have greater interaction with lipid layer, thus can exert high antioxidant activity [21]. Another important consideration could be compounds like eugenol and β-caryophyllene which contains hydrogen atoms in the benzylic and/or allylic positions. These compounds will have better activity in this assay because of relatively easy abstraction of atomic hydrogen from these functional groups by peroxy radicals formed in the test conditions. Another point which favors antioxidant role of eugenol in nutmeg could be that it promotes activities of catalase, superoxide dismutase, glutamine transferase, glutathione peroxidase, and glucose-6-phosphate dehydrogenase enzymes [36].

3.6. Antimicrobial activity of nutmeg extracts

Extracts of nutmeg were evaluated for antimicrobial activity against gram positive (*B. subtilis* and *S. aureus*), gram negative (*P. putida* and *P. aeruginosa*) bacteria and pathogenic fungi (*A. fumigatus*, *A. niger* and *A. flavus*) using disc diffusion method as well their MICs were also determined. It was observed that the extracts of nutmeg used in the present study possess considerable antimicrobial activity against the tested microorganism. Acetone extract of nutmeg has shown the strongest antimicrobial activity than all other extracts of nutmeg used in the study. Diameter of the zone of inhibition and MICs of the various extracts of nutmeg are shown in Table 2. The present study strongly supports that nutmeg seeds have strong antimicrobial activity against important pathogenic bacteria and fungi. Antimicrobial activity of different extracts could be attributed to the occurrence and concentration of various chemical substances present in that extract. Many compounds have been isolated from the nutmegs which are of antimicrobial importance. Narasimhan and Dhake have reported trimyristin and myristic acid as chief antibacterial principles isolated from nutmeg [20]. Three lignans, erythro-austrobailignan-6, meso-dihydroguaiaretic acid and nectandrin-B have been isolated from the methanolic extract of nutmeg which were reported to have antifungal activity [6]. Takikawa et al., have reported antimicrobial activity of nutmeg against entero-hemorrhagic *Escherichia coli* O157 and found that *E. coli* O157 was highly sensitive to β-pinene [32]. Many plant phenolics have been reported to possess antimicrobial activity [24,4]. Some important

Table 2 Antimicrobial activity and MICs of extracts of nutmeg.

Microorganism	Acetone		Ethanolic		Methanolic		Aqueous		Butanol		Standards	
	DI[a]	MIC[b]	DI[a]	MIC[b]	DI[a]	MIC[b]	DI[a]	MIC[b]	DI[a]	MIC[b]	DI[a]	MIC[b]
Bacteria												
Gram Positive											Gentamicin	
B. subtilis	13.4 ± 0.87	31.25	13.6 ± 0.62	31.25	12.9 ± 0.25	31.25	9.7 ± 0.39	62.50	12.4 ± 0.89	62.5	22.1 ± 0.47	7.81
S. aureus	13.8 ± 0.42	31.25	-	-	11.5 ± 0.69	31.25	12.6 ± 0.21	62.50	-	-	23.3 ± 0.62	3.90
Gram Negative												
P. putida	11.2 ± 0.12	31.25	10.9 ± 0.26	31.25	11.1 ± 0.14	31.25	9.4 ± 0.51	62.50	11.0 ± 0.35	62.5	22.7 ± 0.57	7.81
P. aeruginosa	9.3 ± 0.28	62.50	8.9 ± 0.47	62.50	9.4 ± 0.28	62.50	-	-	9.7 ± 0.24	62.5	19.2 ± 0.13	7.81
Fungi											Nystatin	
A. fumigates	12.7 ± 0.25	125.0	12.2 ± 0.45	125.0	11.5 ± 0.19	125.0	9.5 ± 0.18	125.0	9.9 ± 0.41	125.0	14.8 ± 0.73	62.50
A. niger	14.4 ± 0.37	62.50	13.5 ± 0.14	62.50	-	-	10.7 ± 0.29	125.0	10.4 ± 0.11	125.0	19.7 ± 0.51	31.25
A. flavus	10.8 ± 0.46	125.0	10.3 ± 0.63	125.0	10.6 ± 0.52	125.0	8.9 ± 0.17	125.0	-	-	20.4 ± 0.28	31.25

"-" No inhibition zone.

[a] Diameter of zone of inhibition (mm) including disc diameter of 6 mm. Each value is expressed as mean ± SD (n = 3).

[b] μg/ml.

antimicrobial compounds reported in nutmeg are α-pinene, β-pinene, p-cymene, β-caryophyllene and carvacrol [12,17]. α-Pinene and β-pinene (pinene-type monoterpene hydrocarbons) have been reported to have antimicrobial activity and supposed to involve in membrane disruption by the lipophilic compounds [11]. Another important component for antimicrobial activities could be carvacrol. It has been reported that carvacrol can cross cell membranes and penetrate inside the cell where it interacts with intracellular sites critical for antimicrobial activities [8,33]. p-Cymene could also be important component because it is a precursor of carvacrol. It has been reported that p-cymene shows weak antibacterial activity but works synergistically with carvacrol in expanding the membrane which in turn causes destabilization of the membrane [34]. β-Caryophyllene has been reported to have anti-inflammatory and antifungal activities [25]. It has been suggested that antimicrobial activity could be attributed to both major and minor components, it is possible that antimicrobial activity of major components is regulated by the some other minor components as well as these minor components can interact with other components to exert antimicrobial activity [2].

3.7. GC–MS analysis of acetone extract

The chemical composition of the acetone extract of nutmeg was analyzed using GC–MS technique. GC–MS analysis of the acetone extract has led to the identification and quantification of 32 different compounds representing 99.49% of the total extract (Table 3). Sabinene (28.61%), β-pinene (10.26%), α-pinene (9.72%) were found as major compounds which is in accordance with previous work [18,22]. Other important compounds are terpinen-4-ol (5.80%), myristicin (4.30%), limonene (3.76%), γ-terpinene (3.71%), (Z)-p-menth-2-en-1-ol (3.21%), isoeugenol (2.72%), elemicin (2.67%), (E)-p-menth-2-en-1-ol (2.15%), myrcene (2.14%), α-phellandrene (1.84%), p-cymene (1.81%), terpinolene (1.63%) and linalool (1.12%). Difference in the chemical composition of the nutmeg has been reported which could be an attribute of difference in climate, cultivar, soil factor and solvent used for the extraction [18].

Table 3 Chemical composition of acetone extract of nutmeg.

S. No.	Compounds	% Content	RT[a]
1.	α-Thujene	0.27	02.94
2.	α-Pinene	9.72	03.30
3.	Camphene	1.50	03.85
4.	β-Pinene	10.26	04.16
5.	Sabinene	28.61	05.46
6.	Myrcene	2.14	06.67
7.	δ-3-Carene	1.05	06.85
8.	α-Phellandrene	1.84	09.63
9.	α-Terpinene	1.61	09.85
10.	p-Cymene	1.81	10.37
11.	Limonene	3.76	10.79
12.	1,8-Cineole	1.59	11.24
13.	γ-Terpinene	3.71	12.91
14.	trans-Sabinene hydrate	1.02	13.30
15.	Terpinolene	1.63	13.62
16.	Linalool	1.12	14.14
17.	(Z)-p-Menth-2-en-1-ol	3.21	14.87
18.	(E)-p-Menth-2-en-1-ol	2.15	15.55
19.	Terpinen-4-ol	5.80	16.35
20.	α-Terpineol	1.62	18.05
21.	Carvacrol	1.54	18.30
22.	Safrole	0.86	18.40
23.	Eugenol	0.89	18.74
24.	Myristic acid	0.21	18.87
25.	Geranyl acetate	0.19	19.11
26.	Isoeugenol	2.72	19.60
27.	β-Caryophellene	0.82	21.49
28.	β-Cubebene	0.29	21.55
29.	trans-α-Bergamotene	0.31	22.15
30.	Germacrene D	0.27	22.01
31.	Myristicin	4.30	22.61
32.	Elemicin	2.67	23.36
	Total	99.49	

[a] Retention time (min).

4. Conclusions

The present study has demonstrated the antioxidant and antimicrobial activity of various extracts of nutmeg. We have observed that acetone extract has shown the highest antioxidant and antimicrobial properties. High antioxidant and antimicrobial activity could be an attribute of α-pinene, β-pinene, myrcene, 1,8-cineole, carvacrol, terpinen-4-ol, eugenol and isoeugenol. Our study strongly supports the ethno-pharmacological importance of the nutmeg. The antioxidant and antimicrobial activity possessed by nutmeg could be helpful in preventing or slowing the progress of various oxidative stress-related diseases and infections by opportunistic pathogenic microorganisms.

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