Biological Activity and Phytochemical Composition of the Volatile Oils from *Basilicum polystachyon*

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(Received on 7th May 2013, accepted in revised form 21st September 2016)

Summary: This paper extracted and determined the chemical components of the volatile oil in *Basilicum polystachyon*, and measured and evaluated the bioactivity of the volatile oil in *Basilicum polystachyon*. The oils were obtained by hydrodistillation, and their chemical compositions were separated and determined by gas chromatography-mass spectrometry (GC-MS). Minimum inhibitory concentrations (MIC) were determined by using the 8 kinds of plant pathogenic fungi. The free radicals scavenging activity of its volatile oil for the IC_{50} were investigated by using Trolox as the comparison and cytotoxicity by brine shrimp lethal bioassay. The results show that 64 constituents of oils isolated respectively from *Basilicum polystachyon* were identified. The appraised components take up 99.75% of the total peak area. The main composition of the volatile oil is sesquiterpenoids and monoterpene. The results exhibit that the volatile oil in *Basilicum polystachyon* has very strong bioactivity of antimicrobial, antioxidant and cytotoxicity. These results provided the reference for further understanding the chemical components and its bioactivity of this aromatic plant as well as its further development.

Keywords: *Basilicum polystachyon*, Volatile oil, GC-MS, Antimicrobial, Antioxidant, Cytotoxicity

Introduction

Basilicum polystachyon (L.) Moench is a plant belonging to Basilicum genus, Labiatae family [1]. It is widely distributed in the tropical areas in the east hemisphere, including Asia, Africa and Australia. This plant has about six to seven species. In China there is only one specie, which can be seen in Guangdong, Hainan and Taiwan. *Basilicum polystachyon* is an annual or perennial herbaceous plant, and it is a very rare labiatae plant. According to the literature reports, the labiatae plant contains the constituents like terpene, coumarins and volatile oil [2], etc. It has much medicinal effect, such as general anti-bacteria, antivirus, anti-inflammation, analgesia and immune-boosting [3].

Through literature retrieval, there have been no reports about the research on the chemical composition and the bioactivity of *Basilicum polystachyon*. This plant is a kind of aromatic plant which contains much volatile oil. It is widely grown naturally in China, Sanya, Hainan. This paper adopts the hydrodistillation method to extract the chemical elements in the volatile oil in *Basilicum polystachyon*, uses the gas chromatography-mass spectrometry (GC-MS) to determine the chemical composition of the volatile oil ; uses the method of peak area normalization to determine the relative percentage of the volatile oil, uses the prawn larva as the study objects and screens the cytotoxicity of the volatile oil by brine shrimp lethal bioassay

Experimental

Instrument and Materials

Trace MS Gas Chromatograph-Tandem Mass Spectrometer (Amrican Phinigan Corp. Ltd), the chromatographic column is DB-WAX (30 m× 0.25 mm, 0.25 mm) elastic quartz capillary column, Infinite M 200 Microplate Reader (Swiss Tecan Company), UV-2102 PCS Ultraviolet and Visible Spectrophotometer (Shanghai Unica Instrument Corp. Ltd). *Basilicum polystachyon* was picked in Sanya, Hainan in Aug., 2011 and was identified by Professor Huang Shiman, who majored in medicinal plant taxonomy, Hainan University. The chosen strains for the experiment: *Fusarium graminearum, Botrytis cinerea, Exerohilum turcicum, Mucor, Sclerotinia sclerotiorum, Rhizoctonia solani* AG1-IA, *Rhizoctonia solani* Kühn, *Fusarium graminearum* Schwabe. These 8 fungi are provided by the Forestry-Protection Lab, Zhejiang Agriculture and

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Forestry Universtity. The 1,1-Diphenyl-2-picryl- hydrazyl, DPPH, 6-Hydroxy-2,5,7,8- tetramethychroman-2-carboxylic, Trolox are all purchased from Sigma Company, Artemia salina L.eggs are purchased from Fengnian Aquiculture Corp., Tianjin, China; the analytical pure is DMSO.

The Extraction of the Volatile Oil

Weigh 250 g of the plants powder after it being dried, ground and screened through the 20 sieve; then put it into the round-bottomed flask and add a suitable amount of distilled water; the oil was obtained by hydrodistillation for 4 h through the volatile oil extractor according to the XD [4] extracting standard in the appendix of Part One, Pharmacopoeia of the People's Republic of China, 2010 version. Collect the distillate and extract it by diethyl ether. The collected oil was dried with anhydrous Na_2SO_4 and stored at 4°C waiting for analysis and biological activities test. The volatile oil has very rich fragrance. The oil-getting rate of *Basilicum polystachyon* is 2.60%.

The Analytical Conditions of GC-MS

The GC conditions: the Chromatographic column is DB-WAX (30 m× 0.25 mm, 0.25 mm) elastic quartz capillary column; Temperature programming: keep the initial temperature at 45℃ for 3 min, then raise the temperature to 100℃ at the speed of 10℃·min^{-1}, and again raise the temperature to 170℃ at the speed of 5℃·min^{-1}, then again to 240℃ for 7 min at the speed of 10℃·min^{-1}. The temperature at the sample-feeding gate is 250℃, the temperature in the carburetor room is 250℃, the carrier gas is helium; its flow velocity is 0.8 mL·min^{-1}, and the split sampling speed is 20 mL·min^{-1}.

The mass spectrum conditions: Let the electrons bombard the EI ionization source; the ionizing energy is 70 eV; the temperature of the ionization source is 200℃; the voltage of the detector is 350V; the scanning quality range is between 40-300 m./z; the retrieved atlas databank is the standard mass spectrum depot of Willey and NIST; the scanning speed is 0.5 s; the temperature of the quadruple rod is 130℃. Each mass spectrogram corresponding to each chromatographic peak is qualitatively determined by computer chart-base; the relative content of each component is calculated by the Peak Area Normalization method according to its total ion current chart.

RI value

This experiment adopts n-alkane mixed reference sample to analyze according to the GC-MS conditions, and use the Peak Area Normalization method to determine the relative percentage of each chemical constituent in the volatile oil. Then calculate the RI value of each constituent by the linear equation according to the retention time of each n-alkane. RI=100n+ 100 $(t_x- t_n)/t_{n+1}- t_n$. here we analyze and group t_x、t_n and t_{n+1} respectively, with the carbon number as the retention time(min) of the outflow peak of n and n+1 n-alkane (tn< tx< tn+1) [5] .

Bioactivity Determination of the Lethal-to-prawn

The preparation of the sample solution: Weigh precisely 0.02 g of the volatile oil sample; and use DMSO to dissolve it to constant volume 10 mL, and get the sample solution with the concentration of 2 mg mL^{-1}. Then use DMSO to prepare it to the following sample solutions with five different concentration gradient: 10, 50, 100, 500, 1000 μg mL^{-1}. Take the sample solutions with different concentrations and 25-30 prawn larva and experiment it in the 96-hole porous plate. Only add DMSO into the control group, nauplii were used in subsequent experiments after 48 h incubation at in the dark at the room temperature and calculate the number of the dead prawn larva under the microscope in each trough. And finally calculate the mortality rate of the prawn larva according to the equation below: M=(A-B-N)/(G-N)×100%. According to the average death rate under different concentrations calculate its half-number-death concentration LC_{50} by the SPSS method.

The determination of its antimicrobial activity

The slanting test-tube method was adopted to determine the antibacterial activity of the volatile oil and judge its antibacterial ability according to the minimum inhibitory concentration MIC. And then put the extracted volatile oil into the 0.5 mL-1.0 mL flask. Next we mix the 95% ethanol into it till its constant volume by shaking evenly and then use the 95% methanol to dilute it into the concentration gradient with ten different grades 100, 50, 25, 12.5, 6.25, 3.125, 1.563, 0.781, 0.391, 0. 195 , 0.0975 μL mL^{-1}. Inject the prepared volatile oil onto the surfaces of PDA in the test-tube with the same dosage, and the sample-feeding amount each time is 40 μL. After shaking them evenly, add only DMSO on the surface

of the negative control group but add nothing on the surface of PDA in the blank control group. After cultivating in the incubator for 48 h, observe the growth of the fungi and use the minimum concentration of the volatile oil as the minimum inhibitory concentration MIC [6-10].

The determination of the antioxidant activity

DPPH is a free-radical compound and has been widely used to test the free-radical scavenging ability of various samples which has been widely used in evaluating the antioxidant activity of various natural plant extracts. DPPH is a kind of stable free radical in the organic solvents, with its lone pair electrons having the strong absorption at 517 nm. When there is organic scavenger, the lone pair electron will be paired, absorbed, dissolved or weakened. Through measuring their absorption can evaluate the activity of the free radical scavenger [11-16]. With Trolox as the comparison and adopting DPPH method, we measured the antioxidant ability of the volatile oil and calculated its free-radical scavenging ability according to the following equation [17]: Scavenging %=1- (Ap-Ac)/Amax ×100%. In this formula. Use ethanol to dilute the Trolox reserve solution into the different concentration gradients and according to the above method and formula, calculate the Trolox's free-radical scavenging ability for DPPH free radicals. Then we use the fresh working solution to correct it, and regard Trolox concentration as the X-axis and the scavenging rate for the DPPH free radicals as the Y-axis to draw a standard curve. And at the same time, use ethanol to dilute the volatile oil into different concentration gradient, and use the sample concentration as the X-axis and the scavenging rate for the DPPH free radicals as the Y-axis to draw a standard curve.

Results and Discussions

GC-MS analysis result

The volatile oils were a yellow liquid with very rich fragrant smell. The oil yield (v/w) of *Basilicum polystachyon* was 2.60%**.** The components in the volatile oil of *Basilicum polystachyon* and their relative content calculated by the peak area normalization method are their retention indices the mass spectrum database by the NIST2008 standard and combining the artificial analysis. The result was coincident high matching degree with calculate kI value (Fig. 1).

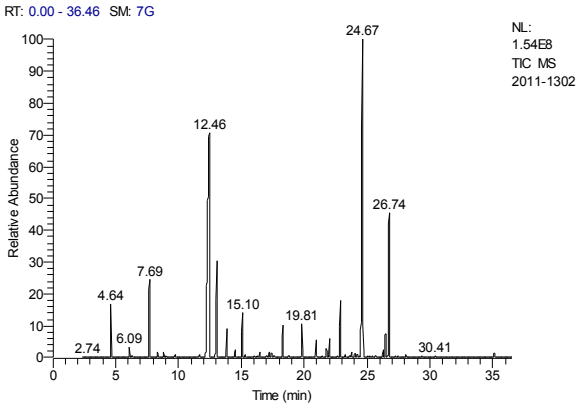

Fig. 1: GC-MS total ion current chromatogram of the essential oil from the *Basilicum polystachyon.*

As can be seen from the Fig. 1, 64 kinds of compounds are determined from the volatile oil of *Basilicum polystachyon*. The determined components take up 99.75% of the total peak area. The highest content of the components in volatile oil are sesquiterpenoids and monoterpene. The relatively higher content of the components in the volatile oil is mainly compound **1**: Ylangene (33.43%), compound **2**: Epiglobulol (31.52%), compound **3**: Copaene (6.14%), compound **4**: Verticiol (5.95%), compound **5**: Caryophyllene oxide (3.01%), compound **6**: D-Limonene (2.93%), compound **7**: Caryophyllene (2.13%), compound **8**: 1R-α-Pinene (1.93%), compound **9**: 1,2,4α,5,8,8α-hexahydro-4,7- dimethyl-1-(1-methylethyl) -[1S-(1α,4αα,8αα)]-naphthalene (1.52%) and its compound structure can be seen in Fig. 2.

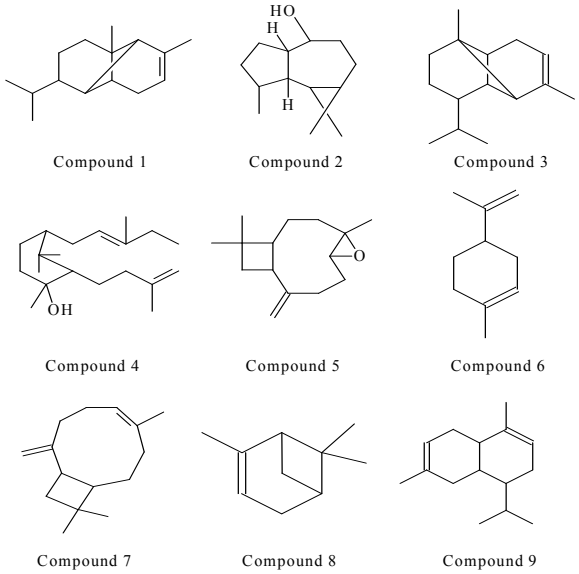

Fig. 2: Representative volatile compounds of essential oil from the *Basilicum polystachyon.*

Table-1: Analytical results of constituents of the essential oil from the *Basilicum polystachyon* by GC/MS.

No.	Name of components	RT (min)	M.F	KI	Relative concent(%)
1	1R-α-Pinene	4.64	$C_{10}H_{16}$	1021.387	1.93
2	4-methyl-1-(1-methylethyl)-didehydro derive bicyclo [3.1.0]hexane	4.71	$C_{10}H_{16}$	1025.434	0.03
3	Camphene	5.34	$C_{10}H_{16}$	1061.850	0.04
4	6,6-dimethyl-2-methylene-(1S)-bicyclo[3.1.1]heptane,	6.09	$C_{10}H_{16}$	1105.263	0.34
5	α-Phellandrene	6.32	$C_{10}H_{16}$	1118.713	0.06
6	4-methylene-1-(1-methylethyl)-bicyclo[3.1.0]hex-2-ene	6.39	$C_{10}H_{14}$	1122.807	0.07
7	D-Limonene	7.69	$C_{10}H_{16}$	1198.830	2.93
8	Eucalyptol	7.85	$C_{10}H_{18}O$	1208.537	0.02
9	2-pentyl-furan	8.17	$C_8H_{14}O$	1228.049	0.03
10	1-Tridecene	8.35	$C_{13}H_{26}$	1239.024	0.16
11	1-methyl-4-(1-methylethyl)-1,4-cyclohexadiene	8.41	$C_{10}H_{16}$	1242.683	0.06
12	1-methyl-2-(1-methylethyl)-b enzene	8.80	$C_{10}H_{14}$	1266.463	0.16
13	1-methyl-4-(1-methylethylidene)-cyclohexene	9.02	$C_{10}H_{16}$	1297.878	0.07
14	1-butenylidene-cyclohexane	9.30	$C_{10}H_{16}$	1296.951	0.03
15	3,4-diethenyl-3-methyl-cyclohexene	9.73	$C_{11}H_{16}$	1321.229	0.11
16	1,3,4,5,6,7-hexahydro-2,5,5-trimethyl-2H-2,4α-ethanonaphthalene	11.62	$C_{15}H_{24}$	1424.742	0.17
17	1-methyl-4-(1-methylethenyl)-benzene	11.73	$C_{10}H_{12}$	1430.412	0.11
18	Thujopsene-I3	12.11	$C_{14}H_{22}$	1450.000	0.07
19	Ylangene	12.46	$C_{15}H_{24}$	1468.041	33.43
20	6-camphenol	12.85	$C_{10}H_{16}O$	1488.144	0.03
21	Copaene	13.07	$C_{15}H_{26}$	1499.485	6.14
22	Octahydro-7-methyl-3-methylene-4-(1-methylethyl)-[3αS-(3αα,3αβ,4α,7α,7αS*)] -1H-cyclopenta[1,3]cyclopropa[1,2]benzene	13.87	$C_{15}H_{24}$	1538.916	1.35
23	(1-propyl-1-nonenyl)-benzene	14.51	$C_{18}H_{28}$	1570.443	0.31
24	1-ethenyl-1-methyl-2,4-bis(1-methylethenyl)-cyclohexane	14.86	$C_{15}H_{24}$	1587.685	0.03
25	Caryophyllene	15.10	$C_{15}H_{24}$	1599.507	2.13
26	Falcarinol (Z)-(-)-1,9-heptadecadiene-4,6-diyne-3-ol	15.31	$C_{17}H_{24}O$	1609.569	0.11
27	1R,3Z,9s-4,11,11-Trimethyl-8-methylenebicyclo[7.2.0]undec-3-ene	16.00	$C_{15}H_{24}$	1642.584	0.17
28	Isoledene	16.28	$C_{15}H_{24}$	1655.981	0.09
29	β-Caryophyllene	16.48	$C_{15}H_{24}$	1665.550	0.22
30	Germacrene D	16.84	$C_{15}H_{24}$	1682.775	0.04
31	ç-Elemene	17.01	$C_{15}H_{24}$	1690.909	0.10
32	1,2,3,4,4α,5,6,7β-octahydro-1,1,4,7-tetramethyl-[1αR-(1αα,4α,4αα,7αβ)]-1H-cycloprop[e]azulene	17.22	$C_{15}H_{24}$	1700.962	0.24
33	(-)-α-Neoclovene	17.26	$C_{14}H_{22}$	1702.885	0.35
34	1,2,3,5,6,7,8,8α-octahydro-1,8α-dimethyl-7-(1-methylethenyl)-, [1S-(1α,7α,8αα)]-naphthalene	17.50	$C_{15}H_{24}$	1714.423	0.08
35	1,2,4α,5,8,8α-hexahydro-4,7-dimethyl-1-(1-methylethyl)-[1S-(1α,4αα,8αα)]-naphthalene	18.30	$C_{15}H_{24}$	1752.885	1.52
36	1,2,3,4,4α,7-hexahydro-1,6-dimethyl-4-(1-methylethyl)-naphthalene	18.76	$C_{15}H_{24}$	1775.000	0.07
37	1,2,3,4-tetrahydro-1,6-dimethyl-4-(1-methylethyl)-(1S-cis)-naphthalene	19.81	$C_{15}H_{22}$	1825.980	1.46
38	3-t-Butyl-4-methoxyphenol methyl derivative	20.43	$C_{12}H_{18}O_2$	1856.373	0.04
39	1-Hydroxy-1,7-dimethyl-4-isopropyl-2,7-cyclodecadiene	20.97	$C_{15}H_{26}O$	1882.843	0.73
40	1,2-dihydro-1,5,8-trimethyl-naphthalene	21.43	$C_{13}H_{16}$	1905.699	0.07
41	Diepi-α-cedrene epoxide	21.77	$C_{15}H_{24}O$	1923.316	0.36
42	1-Hydroxy-1,7-dimethyl-4-isopropyl-2,7-cyclodecadiene	22.03	$C_{15}H_{26}O$	1936.788	0.80
43	Ledene oxide-(II)	22.62	$C_{15}H_{26}O$	1967.358	0.09
44	Caryophyllene oxide	22.88	$C_{15}H_{24}O$	1980.829	3.01
45	4,4α,5,8-tetrahydro-5,8-dimethyl-(4αα,5α,8α)-5,8-epoxy-3H-2-benzopyran	23.25	$C_{11}H_{14}O_2$	2000.000	0.09
46	Decahydro-1,1,4,7-tetramethyl-[1α-(1αα,4α,4αα,7α,7αα,7αβ)]-1H-cycloprop[e]azulen-4-ol	23.62	$C_{15}H_{26}O$	2023.567	0.08
47	1,5,5,8-tetramethyl-[1R-(1R*,3E,7E,11R*)]-12-oxabicyclo[9.1.0]dodeca-3,7-diene	23.77	$C_{15}H_{24}O$	2033.121	0.24
48	Caryophyllene oxide	24.03	$C_{15}H_{24}O$	2049.682	0.44
49	Cubenol	24.20	$C_{15}H_{26}O$	2060.510	0.19
50	Oxacyclotetradeca-4,11-diyne	24.40	$C_{13}H_{20}O$	2073.248	0.37
51	Epiglobulol	24.67	$C_{15}H_{26}O$	2090.446	31.52
52	1,8-Cyclopentadecadiyne	24.95	$C_{15}H_{22}$	2112.264	0.03
53	Decahydro-1,1,7-trimethyl-4-methylene-[1α-(1αα,4αα,7α,7αα,7αβ)]-1H-Cycloprop[e]azulen-7-ol	25.05	$C_{15}H_{24}O$	2121.698	0.07
54	Ledene oxide-(II)	25.21	$C_{15}H_{24}O$	2136.792	0.09
55	.tau.-Muurolol	25.88	$C_{15}H_{28}O$	2200.000	0.03
56	3,3,6,6,9,9-hexamethyl-Z,Z,E-tetracyclo[6.1.0.0(2,4).0(5,7)]nonane,	26.19	$C_{15}H_{24}$	2204.386	0.09
57	4,4,11,11-tetramethyl-7-tetracyclo[6.2.1.0(3.8)0(3.9)]undecanol	26.28	$C_{15}H_{24}O$	2212.281	0.23
58	3,7,11-trimethyl-(Z,E)-2,6,10-dodecatrien-1-ol	26.36	$C_{15}H_{26}O$	2219.298	0.04
59	2-methylene-6,8,8-trimethyl-tricyclo[5.2.2.0(1,6)]undecan-3-ol	26.45	$C_{15}H_{26}O$	2227.193	0.73
60	Verticiol	26.74	$C_{20}H_{36}O$	2252.632	5.95
61	1-ethenyl-1-methyl-2,4-bis(1-methylethenyl)-cyclohexane	27.43	$C_{15}H_{24}$	2314.706	0.07
62	Octahydro-1,4,9,9-tetramethyl-1H-3α,7-Methanoazulene	27.93	$C_{15}H_{26}$	2363.725	0.03
63	3,7,11,15-Tetramethyl-2-hexadecen-1-ol	29.33	$C_{20}H_{40}O$	2500.813	0.06
64	trans-Z-α-Bisabolene epoxide	30.50	$C_{15}H_{24}O$	2595.935	0.04
Total					99.75

The Analysis for the Cytotoxicity Activity

The bioactivity determining method of prawn-larva mortality is a new kind of speedy, concise and practical preliminary screening method for sifting the anti-tumor medicine. The dried prawn larva can survive for several years. At the same time, they can be hatched to many larva at the room temperature when placed in the seawater. Thus, there will be an easy way of getting the prawn larva to be screened without the sterile operation and the animal serum. In addition, this way has the advantage of low cost and small amount of medicine, which is convenient in mass biostatistics. Therefore, it is suitable for screening the cytotoxicity and anti-tumor activity of the plant extracts.

According to the method in the literature [18], we hatch the prawn eggs into the larva, and then put 30 larva into each prepared concentration group (solubilizing the sample by 1% DMSO, and using the artificial seawater solution containing DMSO as the blank control) . Under the room temperature, observe the survival of the larva for 24 h, and then calculate their death rate and the median lethal concentration LC_{50}.

It was reported that the mortality of natural compounds to brine shrimp and their inhibition rate to cancer cells are relativity close, crude extract with LC_{50}<1000 μg·mL^{-1} and pure compound with LC_{50}<100 μg·mL^{-1} had strong cytotoxicity. Through this prawn larva–lethal experiment, it shows that there is no death in the control group, and the death rate is 38% at the concentration of 10 μg mL^{-1}; the death rate is 56% at the concentration of 100 μg · mL^{-1}; the death rate is 100% at the concentration of 1000 μg · mL^{-1}; and the LC_{50} value is 70 μg · mL^{-1}, which shows that the volatile oil of *Basilicum polystachyon* has strong cytotoxicity and anti-tumor activity for the prawn larva.

The analysis for the antibacterial activity

Use the slanting test-tube method to measure the antibacterial activity of the volatile oil. The value of the minimum inhibitory concentration MIC for the 8 kinds of plant pathogenic fungi of the volatile oil in *Basilicum polystachyon* can be seen in Table-2. The experiment results show that the volatile oil of *Basilicum polystachyon* has a clear inhibitory function for the 8 kinds of plant pathogenic fungi. Among them, the highest antibacterial activities appear in *Rhizoctonia solani* AG1-IA and *Rhizoctonia solani* Kühn, and MIC values are both 0.195 μL mL^{-1}; the relatively lower antibacterial activity is for *Fusarium graminearum*, with the MIC 50 μL mL^{-1}.

Recently, the study on the pharmacological activities of eugenol showed that eugenol exhibit diverse activities against the food bacteria. Eugenol can increase the permeability of cell membranes and finally cytoplasmic membrane ruptures. *Fusarium graminearum* exhibited moderate antimicrobial activities. Considering the chemical composition of essential oil, they contain less phenolic components and oxygenation terpenoid, Theresults showed that greater antimicrobial activity potential could be ascribed to the oxygenated terpenes

The analysis for the antioxidant activity

IC_{50} value is an indicator which is often used to evaluate the antioxidant power. It refers to the concentration needed to scavenge 50% DPPH free radicals. The smaller its value, the stronger its scavenging power, and the stronger the corresponding antioxidant power of the tested samples.

We determined their antioxidant activities by DPPH method after extracted the volatile oil of *Basilicum polystachyon* and the artificial antioxidants Trolox into 5 different concentration gradients respectively, and then analyze the results by regression analysis. Then use the concentration of the sample solution to be tested and the concentration of Trolox as the X-axis, and the free-radical scavenging rate (Y) as the Y-axis to establish the standard curve, IC_{50} value was obtained according to the regression equation (Table-3).

The oils of *Basilicum polystachyon* belong to the phenolic compounds. Studies have shown that phenolic compounds play an important role in scavenging free radicals. Mainly due to the redox properties and chemical structure, phenols, secondary metabolites in plant, can play an important role in chelating transition metal, and finally accomplish inhibition of lipoxygenase and scavenging free radicals process.

Table-2: The antifungal activity of the essential oil from the *Basilicum polystachyon* as MIC.

Microorganisms	MIC (μL mL^{-1})	Microorganisms	MIC (μL · mL^{-1})
Botrytis cinerea	25	*Sclerotinia sclerotiorum*	3.125
Exerohilum turcicum	25	*Rhizoctonia solani* AG1-IA	0.195
Mucor	6.25	*Rhizoctonia solani* Kühn	0.195
Fusarium graminearum	50	*Fusarium graminearum* Schwabe	1.563

It is clear in Table-3 that the volatile oil of *Basilicum polystachyon* has a certain function of scavenging the DPPH free-radicals, and its scavenging rate increases with the increase of the concentration of the volatile oil, which shows that there is a positive correlation between the free-radical scavenging rate and the concentration of the volatile oil.

Table-3: Antioxidant activity of the essential oil from the *Basilicum polystachyon* given as IC_{50}.

Sample	Regression equation	R^2	IC_{50}
The essential oil	y =0.4629x + 2.5317	0.9334	0.2710 mL·mL^{-1}
Trolox	y =0.0792x + 0.1948	0.9974	0.1873 mg·mL^{-1}

Conclusion

64 kinds of compounds are determined from the volatile oil of *Basilicum polystachyon*. The appraised components take up 99.75% of the total peak area. The highest content of the compound in the volatile oil is Caryophyllene. This experiment measured the cytotoxicity of the volatile oil in *Basilicum polystachyon* by the method of prawn larva-lethal bioassay. It is reported that when the LC_{50} value of the plant crude extract is less than 1000 μg·mL^{-1}, and the LC_{50} value of its monomeric compound is less than 50 μg mL^{-1}, it will have the strongest cytotoxicity and anti-tumor activity. This experiment also shows that there will be a certain cytotoxicity activity when the LC_{50} value of the volatile oil is 70 μg mL^{-1}.

The results of the antibacterial activity experiment show that the volatile oil of *Basilicum polystachyon* has an obvious inhibitory action for the 8 plant pathogenic fungi. The antibacterial power is the strongest for *Rhizoctonia solani* AG1-IA and *Rhizoctonia solani* Kühn; the antibacterial power is relatively weak for the *Fusarium graminearum.*

The antioxidant activity experiment shows that the IC_{50} value of the volatile oil in *Basilicum polystachyon* for scavenging DPPH free radicals is 0.2710, which shows it has a certain power of antioxidant activity. This *Basilicum polystachyon* is abundant in the natural world and contains much oil as well as very high oil content, so it has a very good prospect in its application and development.

Acknowledgement

We are grateful to Prof. Liping Wang, Analysis and Testing Centre, Jiangnan University, for performing GC-MS, we also thank the microbiology laboratory, Zhejiang Agriculture and Forestry University for performing microorganism experiment, and grateful to The Nurturing Station for the State Key Laboratory of Subtropical Silviculture, Zhejiang Agriculture and Forestry University for performing Universal Microplate Spectrophotometer.

This project is sponsored by Zhejiang Provincial Natural Science Foundation of China (LY13H280011). All authors have declared that no competing and conflicts of interests in above studies.

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