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Chemical Composition of the Essential Oil and the Solvent Extracts of Curcuma Pseudomontana Rhizomes and in Vitro Evaluation of their Antioxidant and Anti-Inflammatory Activities

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Abstract

The phytoconstituents of the essential oil isolated by hydrodistillation and solvent extracts (PE, BZ, DCM, EtOAc and EtOH) of mature rhizomes of turmeric (Curcuma pseudomontana) were analyzed by GC–MS. The major constituents identified in the oil and the extracts were Benzofuran (6.93%), γ-Curcumene (11.18%), β-Elemenone (22.14%), Germacrone (15.15%), Phenol, 2-Methyl-5-(1,2,2-trimethylcyclopentyl)- (29.58%), Germacrene (8.78%), Italicene (15.30%), Xanthorrhizol (56.52%), Pseudocumenol (24.56%) and Epicurzerenolne (4.95%). Analyzing the anti-inflammatory activity of essential oils (EO), and solvent extracts using HRBC stabilization and antioxidant activity. The results of GC MS analysis showed that while Pseudocumenol the major componenets in the essential oil. Xanthorrhizol is the major component in BZ, DCM and EtOH extracts of the matured rhizomes of C. psedomonata. The results of EO and the solvent extracts from Curcuma possessed good anti-inflammatory and antioxidant activities and hence can be used in natural preservative ingredients in food and/ or pharmaceutical industries.

Keywords: **Curcuma Pseudomontana, Essential Oil, Solvent Extracts, in Vitro Anti-Inflammatory, Antioxidant Activity**

Introduction

Turmeric (*Curcuma pseudomontana*) has been used for centuries in Ayurvedic medicine for its anti-inflammatory and antioxidant properties. *C. pseudomontana*, a perennial member of the *Zingiberaceae* family is cultivated mainly in India and South East Asia [1] Recent studies and extensive review of literature has proved the role of curcumin in enhancement of wound healing activity [2-4] It is extensively used as a spice in domestic cooking. In

combination with other natural dyes, it is also used as a coloring agent for textiles, pharmaceuticals, confectionary and cosmetics [5] Essential oils have been reported to be useful in food preservation aromatherapy and fragrance industries. Essential oils are distilled from the leaves, stem, flowers, bark, root and other parts of a given plant botanical typically by steam or water. In citrus fruits, the essential oil is usually pressed from the peels (rinds) of the fruit. There is usually no oily feeling when using essential oils. They are different from vegetable or nut oils because essential oils are not fatty and are highly aromatic. Most of the essential oils are clear, but some oils such as patchouli, orange and lemongrass are amber or yellow in colour. Essential oils contain the true aromatic essence of the plants that they are derived from. Essential oils are highly concentrated and are generally not referred to as scent or fragrance because they offer many other therapeutic benefits that span beyond their pleasant aromas.

The essential oils and the plant extracts have been used for several hundreds of years in food preservation, pharmaceuticals, alternative medicine and natural therapies and it is necessary to scientifically investigate the plants which have been used in traditional medicine to improve the quality of healthcare.

The essential oil extracted from turmeric is reported to possess anti-inflammatory, antifungal, antihepatatoxic and antiarthritic activities and curcumin isolated from it has been reported to possess antioxidative, antimicrobial, antifungal, antiviral and anti-inflammatory properties. The sesquiterpenoids like α-turmerone, β-turmerone and curlone have been shown to possess antioxidative properties Providing protection against oxidative degradation of food is one of the main uses of antioxidants as additives Although many synthetic chemicals, such as phenolic components are found to be potent radical scavengers, they usually have serious side effects and hence antioxidant substances obtained from natural sources will be of great interest now a day.

In the present investigation, we have examined the chemical composition of the essential oil and five solvent extracts of *C. pseudomontana* rhizomes and also evaluated their antioxidant and anti-inflammatory activities by in vitro methods. It would be valuable if such information were available, as antioxidants and anti-inflammatory properties are found in plant extracts and essential oils can be beneficial for maintenance of optimal health and may increase the demand of these bioactive substances in food, cosmetic and pharmaceutical industries

Material and Mehods

Chemicals and Reagents

All the chemicals and solvents used are of analytical grade. Potassium ferricyanide, diphenyl picrylhydrazyl (DPPH) radical, sodium nitro prusside and Diclofenac were purchased from Fluka chemicals and Petroleum ether (PE), benzene (BZ), dichloromethane (DCM), ethyl acetate (EtOAc) and ethanol (EtOH) were purchased from Merck, Mumbai, India

Extraction of the Essential Oil

The mature rhizomes needed for the study were collected from Ootacamund, Tamil Nadu. The plant was authenticated by Dr. R. Kumuthakalavalli, Professor of Biology, Gandhigram Rural Institute, Deemed University, Gandhigram, and Tamil Nadu. Essential oil was extracted from mature rhizomes of *C. pseudomontana* (500 g) by hydro distillation using a Clevenger's apparatus for 3 h and the extracts were prepared by extraction of 500 g of the rhizomes each with petroleum ether, benzene, dichloromethane, ethyl acetate and alcohol as solvents individually for 6 h. The yellow coloured essential oil collected was dried over anhydrous sodium sulphate and stored at 40 C for further use. We also stored individual solvent extracts for further use in a refrigerator after removing the solvents.

Gas Chromatographic Analysis (GC/MS) of the Oil and the Solvent Extracts

The GC-MS analysis of the EO was performed on an Agilent GC 6890N model gas chromatograph-5973N model mass spectrometer equipped with a 7683B series auto-injector (Agilent, USA). The GC was equipped with a HP-5MS Capillary Column (30 m x 0.25 mm x 0.25 µm film thickness). The column temperature program began at 40 ºC and was held for 3 min. It was increased at a rate of 10 ºC min^{-1} to 150 ºC and held for 8 min and then increased at a rate of 10 ºC min^{-1} to 280 ºC and held for 15 min. Injection volume was 1.0 ML and inlet pressure was 7.06 psi. Helium was used as the carrier gas and linear velocity (u) was 36 cm/sec. Injection mode was split (30:1) and MS interface temperature was 250°C. Mass spectra were recorded in the scan mode at energy was 70 eV and MS spectra were scanned from 50 to 550 m/z at 2.2 scan s^{-1}. With the help of a MS library, compounds were identified by comparing retention indices. The NIST 05a and Wiley7a spectrometer data bank was used to determine the percentage composition of the compounds [18].

DPPH Radical Scavenging Assay

The stable 1,1-diphenyl-2-picryl hydrazyl (DPPH) was used for the determination of the free radical scavenging activity of the EO and the solvent extracts by the method suggested by [19]. Test solutions of different concentrations (5,10,15, 20 and 25 µl/mg) of the EO, solvent extracts, and the standard gallic acid were separately added equal volume of 100 mM DPPH solution. A UV-visible spectrophotometer was used to measure each reaction mixture's absorbance at 517 nm after 30 minutes at room temperature. The free radical scavenging activity of the test samples was calculated using the formula

Percentage of Free Radical Scavenging activity = $[(A_{control} - A_{sample}) / A_{control}] \times 100$

where $A_{control}$ is the absorbance of the blank and A_{sample} is the absorbance of the sample solution.

Hydrogen Peroxide Scavenging Assay

A solution of H2O2 (40 mM) was prepared in phosphate buffer (pH 7.4). Stock solutions, each containing 1 mg/ml of the EO and the solvent extracts were prepared. Different amounts (5, 10, 15, 20 and 25 µl/ mg) of the stock solutions taken in different test tubes were added 1 ml of H2O2 solution (40 mM) and the volume was adjusted to 25 ml with Phosphate buffer solution (PBS) and all the reaction mixtures were incubated at 230 nm. By using the same method without the sample, a blank solution was prepared. The percentage of H2O2 scavenging ability of the sample extract or positive control was calculated by using the equation

H_2O_2 scavenging ability % = $[(A_b - A_s)] / A_b \times 100$

where A_b is the absorbance of the blank, A_s is the absorbance of the sample solution.

Reducing Power Assay

The reducing power of the essential oil and the solvent extracts was determined according to the method suggested by [20]. The EO and the solvent extract samples of various concentrations (5, 10, 15, 20 and 25 µl/ mg) in methanol were mixed individually with 2.5 ml of PBS (0.2 M, pH 6.7) and 2.5 ml of potassium ferricyanide (1 %, w/v). The mixture was incubated at 50 0C for 20 min. Trichloroacetic acid (2 ml of 10 %, w/v) was added to the reaction mixture which was then centrifuged for 15 min. The upper layer of solution (2.5 ml) was mixed with pure water (2.5 ml) and ferric chloride (0.5 ml, 0.1 %, w/v), and the absorbance was measured at 700 nm.

Nitric Oxide Radical Scavenging Activity

Stock solutions of the EO and the individual solvent extracts were prepared to contain 1 mg/ml

of the test sample and different amounts (5, 10, 15, 20 and 25 µl/mg) of the stock solution were transferred to different test tubes and the volume was adjusted to 25 ml by the same solvent. 0.2 ml of sodium nitro prusside (20 mM) in PBS (pH 7.4) and 1.8 ml of PBS solution were added and the mixture was incubated at 37 oC for 3 hrs. 1 ml of each solution was diluted with 1 ml of Griess reagent (1% sulfanilamide, 2% H3PO4, 0.1 % N-1-naphthyl ethylene diamine). Similarly a blank was performed containing the equivalent amount of reagents but without the test samples. The absorbance of these solutions was measured at 540 nm against the corresponding blank solutions. Ascorbic acid was used as a positive control (25 µl/mg).

Cupric Ion Reducing Antioxidant Capacity (CUPRAC Assay)

CUPRAC method is a novel hydroxyl radical scavenging antioxidant activity assay for water-soluble antioxidants. Reactive oxygen species (ROS) may attack biological macromolecules giving rise to oxidative stress-originated diseases. Since OH is very short-lived, secondary products resulting from OH attack to various probes are measured. 1 ml each of 10 mM CuC_{l2}, 75 mM neocuproine and NH_4Ac buffer (1 ml, pH 7.0) solutions were taken in a test tube. Then different Concentrations of EO extract, solvent extract and the standard of 5-25 µg / ml were mixed and the total volume was brought upto 40 ml with deionized water.

Human Red Blood Cell Membrane Stabilization Method (HRBC Method)

HRBC stabilization method was used for the estimation of in-vitro anti-inflammatory activity [21] Blood collected from healthy human volunteers was mixed with equal volume of sterilized Alsever solution (2 % dextrose, 0.8% sodium citrate, 0.5% citric acid and 0.42% sodium chloride) and solution was centrifuged at 3000 rpm for 15 min and the packed cells were separated. The packed cells were washed using isosaline solution and a 10% v/v suspension was made with isosaline. This HRBC suspension was used for the estimation of anti-inflammatory property. Different concentrations (5, 10, 15, 20 and 25 µl/mg) of the extract, reference sample and control were separately mixed with 1 ml of phosphate buffer, 2 ml of hyposaline and 0.5 ml of HRBC suspension. The samples were incubated at 37oC for 30 minutes and centrifuged at 3000 rpm for 2 min. The supernatant liquids were decanted and the hemoglobin content of each of them was estimated by using a spectrophotometer at 560 nm. Diclofenac was used as the standard and a control was prepared omitting the test solution. The percentage hemolysis was estimated by assuming the hemolysis produced in the equation

Percentage of Inhibition = (1 – (OD $_{Sample}$ / OD $_{control}$)) x 100

Result and Discussion

Chemical Compositions of the Essential Oil and Solvent Extracts of *C. pseudomontana*

The pale yellow coloured essential oil was obtained by hydro-distillation of the mature rhizomes of the plant and all the solvent extracts of PE, BZ, DCM, EtOAc and EtOH obtained were also light yellow in colour. The chemical compositions of the essential oil and all the

Table 1 Chemical Composition of the Essential Oil and the Solvent Extracts of *Curcuma Pseudomontana*

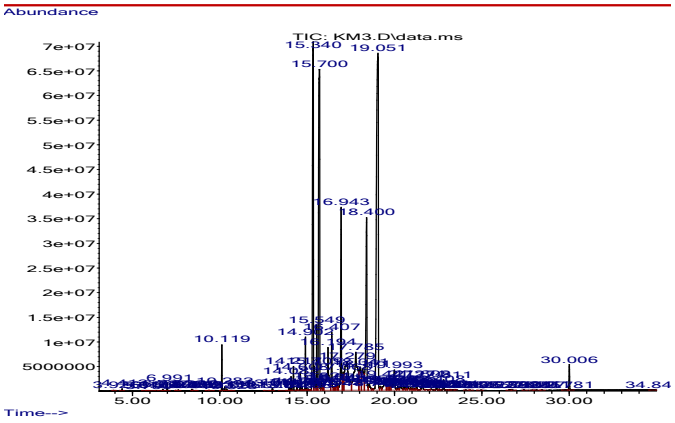
Compound Name	Area peak (%)					
	PE	BZ	DCM	EtOAc	EtOH	EO
Benzene1-(1,5 dimethyl-4-hexenyl)-4-methyl-	9.92	12.52	13.04	13.55	0.56	7.27
Benzofuran	6.50	6.09	6.93	6.78	0.43	2.89

Gamma-Curcumene	11.18	1.04	0.76	0.56	4.53	
Germacrene	3.88	1.4	2.82	5.17	8.78	1.83
3-Ethyl-6-(methoxycarbonyl)-naphthol	6.09	0.23	0.23	-	-	-
3,7-Cyclodecadien-1-one	1.55	-	0.12	1.62	-	-
β-Elemenone	7.93	7.08	0.34	2.01	0.98	22.14
Germacrone	8.25	7.31	7.19	6.45	0.56	15.15
Phenol,2-methyl-5-(1,2,2-trimethylcyclopentyl)-	29.58	0.89	0.13	-	-	-
Isopropyl-1,2,4-trimethylbenzene	1.38	1.77	0.45	0.99	1.23	1.55
2,2'-Butenyl-mesitylene	0.99	1.88	0.14	-	-	-
Isospathulenol	0.58	-	0.52	-	0.23	-
Trans-β-Farnesene	0.58	0.54	0.85	0.79	0.56	-
Camphor	0.44	-	0.42	0.58	-	-
Trans-Caryophllene	1.02	0.39	0.32	0.94	-	0.17
Bicyclo[2.2.1]heptan-2-one	0.21	1.89	0.74	0.45	0.99	0.66
Trans-β-Elemene	0.16	0.78	7.20	5.65	2.98	0.95
Italicene	-	14.50	14.99	15.30	-	-
ϒ-1-Cadinene	-	0.39	-	-	0.54	-
Bicyclogermaacrene	-	2.64	3.14	0.54	-	-
Sativene	-	0.28	0.24	-	-	-
Epicurzerenolne	0.52	4.16	4.95	4.29	3.09	-
Xanthorrhizol	-	27.31	25.35	-	56.52	-
α-Terpinene	-	0.28	1.40	0.23	-	-
Cyclohexanolne	0.07	-	1.95	0.45	-	-
Propanolic acid	-	-	0.56	0.72	-	-
Pseudocumenol	-	-	0.76	24.56	-	20.65
Bis(2-ethyl hexyl)phthalate	-	-	0.64	1.38	-	-
7-Methanol azulene	-	-	0.98	-	4.99	-
1,2-Diethylidene cyclopentane	0.23	-	0.43	-	2.08	-
2-(4-Methoxy phenyl)N,N trimethyl-1-pyrrolamine	-	-	-	-	-	13.12
Allospathulenol	-	-	-	-	-	2.69
5-Ethyl-1,3-dimethylindan	-	-	-	-	-	1.98
1,6-Heptadiene	-	-	-	-	-	1.39

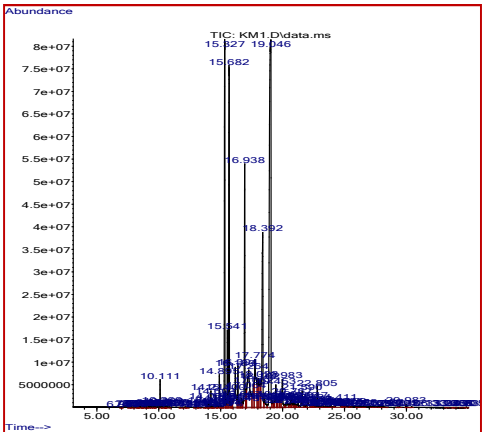
Solvent extracts identified are given in Table 1. The GC-MS analytical study of the essential oil has resulted in the identification of 104 (including supplementary data) components representing 99.43% of the total oil. The major components of the EO were Benzene (1,5 dimethyl-4-hexenyl)-4-methyl- (7.27 %), β-Elemenone (22.14%), Germacrone (15.15 %), Pseudocumenol (20.65 %) and 2-(4-methoxy phenyl)N,N trimethyl-1-pyrrolamine (13.12 %) (Table 1).

The GC–MS analysis of the solvent extracts (PE, BZ, DCM, EtOAc and EtOH) from mature rhizomes of *C. pseudomontana* led to identification of 117, 107, 97, 127 and 51 phytoconstituents respectively in them (as shown in spectra data for GC-MS) representing 96.95% of the total amount of the oil extracted.

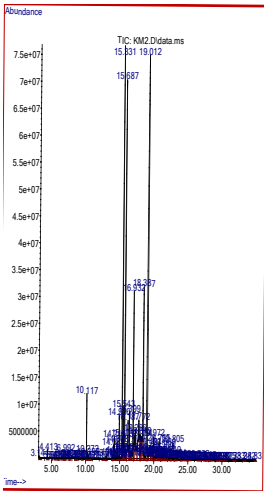
Spectra Data 1 Percentage of the Components of Petroleum Ether Extract of Curcuma Pseudomontana Rhizomes



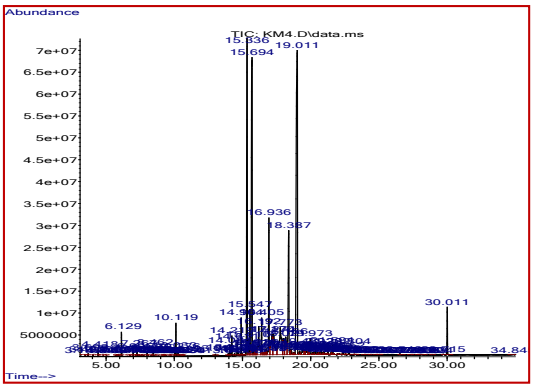
Spectra Data 2 Percentage of the Components of the Benzene of Curcuma Pseudomontana Rhizomes



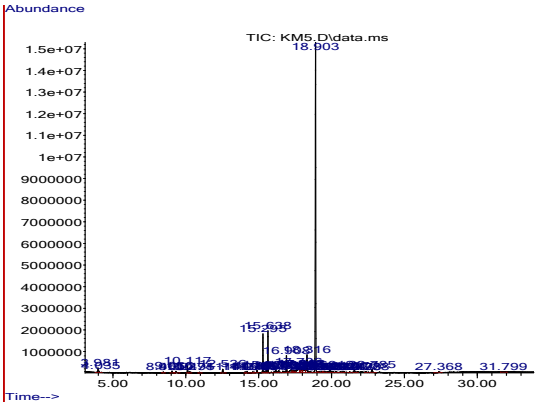
Spectra Data 3 Percentage of the Components of Dichloromethane Extract of Curcuma Pseudomontana Rhizomes



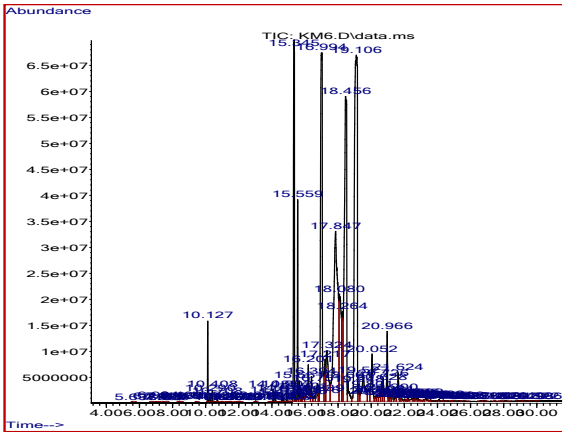
Spectra Data 4 Percentage of the Components of Ethyl Acetate Extract of Curcuma Pseudomontana Rhizomes



Spectra Data 5 Percentage of the Components of Ethanol Extract of Curcuma Pseudomontana Rhizomes



Spectra Data 6 Percentage of the Components of the Essential Oil from Curcuma Pseudomontana Rhizomes



The identified compounds with their retention time and their percentage are also listed in Table 1 where in the area peak are arranged in the order of their retention time. The essential oil was identified to be a complex mixture of mainly oxygenated monoterpenes, sesquiterpenes and hydrocarbons. The major components in the solvent extracts of mature rhizomes of

C. pseudomontana were Benzene1-(1,5 dimethyl-4-hexenyl)-4-methyl (13.55%), Benzofuran (6.93 %), Gamma-Curcumene (11.18 %), β-Elemenone (22.14 %), Germacrone (15.15%), Phenol, 2-methyl-5-(1,2,2-trimethyl cyclopentyl)- (29.58%), Germacrene (8.78%), Trans-β-Elemene (7.20%), Italicene (15.30 %), Xanthorrhizol (56.52 %), Pseudocumenol (24.56 %), Epicurzerenolne (4.95 %), 3-Ethyl-6-(methoxycarbonyl)-naphthol (6.03 %), 7-methanolazulene (4.99%) of mature rhizomes of *C. pseudomontana*.

Antioxidant Activities of the EO and the Solvent Extracts

The role of antioxidants in removing free radical is by donating hydrogen to free radicals there by converting them into unreactive species. Addition of hydrogen would remove the odd electron feature of the reduced responsible for the radical reactivity.

Table 2 In–vitro Antioxidant Assay of the EO, Solvent Extracts and the Standard by *C. Pseudomontana* by DPPH, H2O2, Reducing Power and Nitric Oxide Methods

Test sample	Concentration (µl/mg)	% inhibition				
		DPPH Method	H2O2 Method	Reducing power	Nitric Oxide method	Standard Drug
PE	5	80.2	71.3	78.3	75.6	85.2
	10	81.4	73.2	79.7	76.2	86.3
	15	82.6	74.7	80.5	77.9	87.6
	20	83.3	75.2	81.2	78.7	88.5
	25	84.2	76.5	82.3	79.5	89.3
BZ	5	79.5	79.5	81.2	78.2	80.7
	10	80.2	80.2	82.9	79.5	81.6
	15	81.9	81.6	83.7	80.2	82.3
	20	82.7	82.3	84.6	81.5	83.7
	25	83.4	83.5	85.3	82.6	84.6
DCM	5	85.3	80.6	87.6	80.3	82.7
	10	86.1	81.3	88.5	81.7	83.4
	15	87.5	82.7	89.2	82.6	84.8
	20	88.2	83.5	90.2	83.9	85.2
	25	89.6	84.8	91.4	84.8	86.9
EtOAc	5	83.4	73.5	75.9	76.3	89.5
	10	85.1	74.7	76.7	77.8	90.2
	15	86.3	75.9	77.5	78.2	91.3
	20	87.5	76.8	78.7	79.5	92.7
	25	88.4	78.1	79.8	80.6	93.5
EtOH	5	87.3	82.3	80.2	82.3	90.5
	10	88.7	83.5	81.9	83.4	91.4
	15	89.5	84.7	82.7	84.7	92.8
	20	90.2	85.9	83.8	85.3	93.6
	25	91.5	872	84.7	86.2	94.7

EO	5	90.2	89.6	82.5	89.5	92.4
	10	91.5	90.3	83.7	90.2	93.6
	15	92.7	91.5	84.4	91.5	94.7
	20	93.4	92.6	85.6	92.7	95.9
	25	95.2	93.7	86.9	93.6	96.2

The hydrogen-donating activity, measured using DPPH as hydrogen acceptor, showed that a significant association could be found between the concentration of EO/ the solvent extracts and percentage of inhibition of free radicals at different concentrations ranging from 5 to 25 µl/mg for the EO and the solvent extracts using ascorbic acid as the standard and the results are shown in Table 2.

In reducing power assay, the presence of antioxidants in samples would result in the reduction of Fe^{3+} to Fe^{2+} and the amount of Fe^{2+} complex was monitored by measuring the formation of blue colour at 700 nm. Increase in absorbance at 700 nm indicated an increase in reductive ability. The reducing ability of the essential oil from mature rhizomes of *C. pseudomontana* is shown in Table 2 As discussed in the case of antioxidant activities, the results of study of the essential oil, the solvent extracts and the standard at concentrations of 5, 10, 15, 20, and 25 µl/ mg, showed that the increase in absorbance of the reaction mixture showed an increase in the reducing power indicating the essential oil was having a lesser reducing power than the standard. It is evident from Table 2 that the reducing powers of the essential oil, solvent extracts and also the standard were increasing with high concentration.

H_2O_2 scavenging activity is a measure of the antioxidant activity of the EO and the solvent extracts. In this direct method of involving UV spectrophotometry, the determination is based on the intrinsic absorption of H_2O_2 in the UV region at 230 nm, solutions of all concentrations of the EO and the solvent extracts scavenged hydrogen peroxide which may perhaps be attributed to the presence of phenolic groups that could donate electrons to hydrogen peroxide there by neutralizing them into water. Many plants have often been reported to exhibit antioxidant activity due to the presence of polyphenolics and curcumin as antioxidants [22].

The Nitric Oxide radical scavenging activities of the EO and the solvent extracts and the standard are also shown in Table 2. In the present study, nitric oxide generated from sodium nitro prusside, at physiological pH 7.4 liberates nitrate which gets converted to nitrite which further forms nitrite ions on contact with air. The nitrite ions when diazotized with sulphanilic acid and coupled with naphthylethylenediamine formed the pink color complex, which was measured at 546 nm. The study showed that the antioxidant activity of the EO and the solvent extracts increased dose dependently and the % of inhibition for the EO was slightly less than that for the standard.

In-vitro Anti-inflammatory Study of the EO and the Solvent Extracts

In the present study, the EO and the solvent extracts and the standard at different concentrations (5, 10, 15, 20 and 25 µg/ml) showed significant stabilization towards HRBC membranes at all concentrations dose dependently and the percentage inhibition of the EO and the solvent extracts at a concentration of 25 µg/ml was found to be the maximum and smaller than that of the standard. Except in the case of EtOH extract wherein it was found to be higher than that of standard drug Diclofenac sodium showing an inhibition of (91.0 %) the results are shown in Table 3.

Table 3 In-vitro Anti-inflammatory Activity of the EO, Solvent Extracts of C. Pseudomontana and the Standard by HRBC Membrane Stabilization Method

S.No	Test sample	Concentration (µl/mg)	% inhibition	
			HRBC Method	Standard Drug
1	PE	5	82.7	85.4
		10	83.6	86.6
		15	84.8	87.3
		20	85.2	88.5
		25	87.5	89.9
2	BZ	5	78.5	79.3
		10	79.2	80.2
		15	80.5	82.4
		20	82.3	83.6
		25	83.5	84.8
3	DCM	5	72.5	87.2
		10	73.6	88.6
		15	74.2	89.7
		20	75.3	90.4
		25	76.2	91.7
4	EtOAc	5	82.7	82.2
		10	83.9	83.4
		15	84.7	84.7
		20	85.5	85.8
		25	86.9	86.9
5	EtOH	5	90.2	86.4
		10	92.4	87.2
		15	93.8	88.5
		20	94.7	89.7
		25	96.2	91.0
6	EO	5	89.4	92.3
		10	90.5	94.1
		15	91.8	95.4
		20	92.9	96.5
		25	95.3	97.2

Conclusion

The antioxidant and anti-inflammatory properties of the essential oil and solvent extracts of plants are of great interest in food, cosmetic and pharmaceutical industries and they might be suitable as preservatives to control food-borne pathogens and as natural antioxidants to reduce oxidative stress in human beings. The in-vitro antioxidant study carried out for the oil from the mature rhizomes of *C. pseudomontana* showed maximum inhibition efficiency for the EO against free radicals, which is very nearly equal to that of standard Ascorbic acid (96.2%) at a concentration of 25 µl/mg. The

in-vitro anti-inflammatory study of the oil sample when carried out by HRBC- Method showed maximum inhibition efficiency of EO 95.3% (when compared with 97.2% of inhibition for the standard), which is marginally higher than that for the standard Diclofenac sodium. The results showed that the inhibition efficiency increases with increase in concentration of the test samples and the results are slightly lower than that of the standard

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References

1. H.P. Ammon and M.A. Wahl, Pharmacology of Curcuma longa. Planta Med, 57, 1– 7 (1991).
2. M. Deters, C. Siegers, W. Hansel, K.P. Schneider and G. Hennighausen, Influence of curcumin on cyclosporin-induced reduction of biliary bilirubin and cholesterol excretion and on biliary excretion of cyclosporin and its metabolites. Planta Med,66, 429–434 (2000).
3. B.T. Kurien, and R.H. Scofield, Increasing aqueous solubility of curcumin for improving bioavailability. Trends Pharmacol. Sci, 30, 334–335 (2009).
4. G. Singh, I.P. Kapoor, P. Singh, C.S. de Heluani, M.P. de Lampasona, and C.A. Catalan, Comparative study of chemical composition and antioxidant activity of fresh and dry rhizomes of turmeric (Curcuma longa Linn.). Food Chem. Toxicol, 48, 1026–1031, (2010).
5. G. Singh, I.P.S. Kapoor, S.K. Pandey, and O.P. Singh, Curcuma longa – chemical, antifungal and antimicrobial investigations of rhizome oil. Indian Perfumer, 47, 173–178 (2003).
6. G. Sandri, J. Zacaria, F. Fracaro, A.P.L. Delamare, and S. Echeverrigaray, Antimicrobial activity of the essential oils of Brazilian species of the genus Cunila against food borne pathogens and spoiling bacteria. Food Chem, 103, 823–828 (2007).
7. M.P. Buttner, K. Willeke, and S.A. Grinshpun, Sampling and analysis of airborn microorganisms. Manual of Environmental Microbiology. A.S.M. Press, Washington, DC, 629–640 (1996).
8. S.A.A.J. Van de Braak, G.C.J.J. Leijten, Essential oils and oleoresins: a survey in the Netherlands and other major markets in the European Union. CBI, Centre for the Promotion of Imports from Developing Countries. Rotterdam. 116 (1999).
9. S. Murkas, M. D. Mueller, and E. Mechler, Medicinal plants in tropical countries. Thiem, 5, 168-173 (2005).
10. S. Burt, Essential oils: their antibacterial properties and potential applications in foods – a review. Int. J. Food Microbiol, 94, 223–253 (2004).
11. M. Lis-Balchin, and S.G. Deans, Bioactivity of selected plant essential oils against Listeria monocytogenes. J. Appl. Bacteriol, 82,759–762 (1997).
12. C. Behura, P. Ray, C.C. Rathi, R.K. Mishra, O.S. Ramachandraiah, and J.K. Charyulu, Antifungal activity of essential oil of Curcuma longa against five rice pathogens in vitro. J. Essential Oil Bearing Plants, 3, 79–84 (2000).
13. K. Palasa, B. Scsikaran, T.P. Krishna, and K. Krishnaswamy, Effect of turmeric on urinary mutagens in smokers. Mutagenesis, 7, 107–109 (1992).
14. M.R. Bower, H.S. Aiyer, Y. Li, and R.C. Martin, Chemoprotective effects of curcumin in esophageal epithelial cells exposed to bile acids. World J. Gastroenterol, 16, 4152–4158 (2010).
15. G.K. Jayaprakasha, B.S. Jena, P.S. Negi, and K.K. Sakariah, Evaluation of antioxidant activities and antimutagenicity of turmeric oil: a byproduct from curcumin production. Naturforsch, 57, 828–835 (2009).

16. I. Gulcin, S. Beydemir, I.G. Sat, M. Elmastas, and O.I. Kufrevioglu, Comparison of antioxidant activity of clove (Eugenia caryophylata Thunb.) buds and lavender (Lavendula stoechas L.). Food Chem. 87, 393–400 (2004).
17. K. Imaida, S. Fukushima, T. Shivai, M. Ohtani, K. Nakanishi, and N. Ito, Promoting activities of butylated hydroxyanisole and butylated hydroxytoluene on 2-stage urinary bladder carcinogenesis and inhibition of c-glutamyl trans peptidase-positive foci development in liver of rats. Carcinogenesis, 4, 885–889 (1983).
18. R.P. Adams, Identification of Essential Oil Components By Gas Chromatography/Mass Spectrometry, 4th edition. Allured Publishing Corporation, Carol Stream, Illinois. (2007).
19. I.I. Koleva, T.A. van Beek, J.P.H. Linssen, and A. De Groot Evstatieva, Phytocheml Analysis, 13, 8-17 (2002).
20. G.C. Yen, and H.Y. Chen, Antioxidant activity of various tea extracts in relation to their antimutagenicity. J. Agr. Food Chem, 43, 27–32 (1995).
21. Kim, Jin, Lee, Chang-Moon, Jeong, Hwan-Jeong, Kim, Dong-Woon, Lee, and Ki-Young, J. Nanosci. Nanotechn, 12, 2672-2678 (2012).
22. C.A. Rice-Evans, N.J. Miller, P.G. Bolwell, P.M. Bramley, and J.B. Pridham, The relative antioxidant activities of plant-derived polyphenolic flavonoids. Phytochem, 37, 375-383 (1995).