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Comparative phytochemical screening of *Curcuma angustifolia*, *Curcuma decipiens* and *Curcuma longa* by using GC-MS

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Abstract

Present research was constructed to find out the phytochemical compounds of methanol, ethanol and acetone extracts of *Curcuma longa*, *Curcuma angustifolia* and *Curcuma decipiens*. This investigation also reveals the comparison of methanolic extract of these selected species of family zingiberaceae by gas chromatography and mass spectrometry technique. The main goal of this study to compare phytochemicals of cultivated species with wild species of genus *Curcuma*. The results obtained from GC-MS indicate that 34 phytochemicals were found in *Curcuma angustifolia*, 63 phytochemicals were found in *Curcuma decipiens* and 53 phytochemicals were found in *Curcuma longa*. 10 phytochemical compounds of *Curcuma longa* were identical with *Curcuma angustifolia* and 14 with *Curcuma decipiens*, while other phytochemicals of these wild species were different from *Curcuma longa* and can be used to treat various diseases.

Keywords: curcuma, phytochemical, GC-MS, soxhlet extraction, methanol

Introduction

Plants are excellent source of medicinal agent for many years. This has lead to an increasing curiosity in the research of different extracts from plants as possible source of new antimicrobial agents. Natural medicines have become more well-liked in the treatment of many diseases due to popular faith that these phyto-medicines are harmless, easily accessible, and low cost and having a lesser amount of adverse effects. Many plant medicines are cheaper and more available to most people rather than conventional medicines. There are lower rate of side effects after use. These reason might account for their worldwide attention and use [1]. Several researchers have documented medicinal properties of several plants in different periods. Medicinal plants constitutes the main source of new pharmaceutical and healthcare products [2]. Extraction and characterization of some active phytochemicals from these green factories have given birth to some high active profile drugs [1]. These information of phytoconstituents from the plants are desirable because such information will be significant for the synthesis of chemical substances. Screening of phytochemicals is documented by many researchers as mentioned below.

In manner to study comparative phytochemical analysis of genus *Curcuma*, three species were taken- *Curcuma longa* is a cultivated species (turmeric), *Curcuma angustifolia* (tikhur) and *Curcuma decipiens* (van haldi) are wild species. All these species of curcuma are herbs that possess rhizomes and categorized in family zingiberaceae. Most of the member in this family are rich in phytochemicals and well-known as conventional medicines for various diseases. Curcumin and its derivatives and other extracts from the leaves and rhizomes are bioactive in nature. The chief activities of these extracts have been found to be anti-inflammatory, anti-cancer, antibacterial, antifungal, antiviral and curing many disorders.

This study involves the application of different techniques like Gas chromatography, Mass spectrometry analysis for phytochemical characterization of the herbal drug synthesis.

Materials and Methods

Collection of plants

Three selected species of family zingiberaceae were collected from different areas of India. *Curcuma longa* is a cultivated species so it was collected from a farm of Indore region (Madhya Pradesh). *Curcuma deciepiens* is a wild species which was collected from a forest of kalibhit located in khandwa district (Madhya Pradesh). *Curcuma angustifolia* is also a wild species but now it is also cultivated for medicinal purpose in some areas. So it was collected from Ambikapur (Chhattisgarh).

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Preparation of plant extract

After completion the complex procedure of plants collection, rhizomes were separated from their leaves and peeled and washed properly. Rhizomes of *C. angustifolia* and *C. longa* were sliced into pieces due to bigger size while rhizomes of *C. decipiens* was dried directly. Drying procedure was conducted in shade at ambient temperature then dried rhizomes were pulverized into powder and stored in clean air tight containers until further use.

Preparation of extract by soxhlet extractor

Dried powders were subjected to soxhlet extraction for preparing the extracts. Methanol, ethanol and acetone were used as solvents.

The dried powder of each sample (35gm) was loaded in the thimble of Soxhlet apparatus. 400 ml of solvents were used one by one with each sample. The extraction was continued till clear solvent was seen in the thimble. It takes about 25 hours. Then the concentrated extract was evaporated at normal temperature till a dark coloured residue was obtained. So 9 extracts were prepared with same method. Weighed extracts were kept in air tight containers for preliminary phytochemical analysis. Methanolic extracts of all three plants were analyzed by gas chromatography mass spectrometry technique.

Preliminary phytochemical analysis

These extracts were tested in order to find out the presence of bioactive compounds by use of following standard methods^{4,8}

Test for alkaloids

Wagners test: 1 ml extract was treated with Wagner's reagent; formation of brown reddish precipitate indicates presence of alkaloids.

Test for flavanoids

NH₄OH test: 3 ml of extract was treated with 10 % NH₄OH solution development of yellow fluorescence indicates positive test.

Zn test: 2 ml extract was treated with Zn dust and conc. HCl development of red colour indicates presence of Flavonoid.

Test for Carbohydrates

Extract were dissolved individually in 5ml of distilled water and filtered. The filtrate was used for the following test.

Molisch's Test: Filtrate were treated with 2 drops of alcoholic α -naphthol solution, formation of violet ring at the junction indicates the presence of carbohydrate.

Iodine Test: 2ml of extract was treated with 5 drops of Iodine solution, gives blue color indicates the positive test.

Fehling Test: 2ml of extract was hydrolyzed with dilute HCl and neutralized with alkali & heated with Fehling's solution A and B, formation of red ppt indicates the presence of reducing sugar.

Test for proteins

Xanthoproteic test: Extract was treated with few drops of concentrated HNO₃ formation of yellow indicates the presence of proteins.

Biuret test: Add 2ml of Biuret reagent to 2ml of extract. Shake well and warm it on water bath. Appearance of red or violet colour indicates presence of proteins.

Test for saponins

5 ml extract was mixed with 20 ml of distilled water then agitated in graduated cylinder For 15 min formation of foam indicates Saponin.

Test for phenols tannins

Crude extract was mixed with 2ml of 2% solution of FeCl₃. A blue-green or black coloration indicated the presence of phenols and tannins.

Test for steroids

1ml extract was dissolved in 10 ml of chloroform & equal volume of concentrated H₂SO₄ acid was added from the side of test tube. The upper layer turns red and H₂SO₄ layer showed yellow with green fluorescence. This indicates the presence of steroid.

Test for fatty acids and oils

Spot test: Prepared spot on the filter paper with the test solution and oil staining on the filter paper indicated the presence of fixed oil & fats.

Test for amino acids

Ninhydrin test: To the 2 ml extract 2 ml on ninhydrin reagent was added & boil for few minutes, formation of blue colour indicates the presence of amino acid.

Test for anthocyanin

2 ml of aqueous extract is added to 2 ml of 2N HCl & NH₃, the appearance of pink red turns blue violet indicates presence of Anthocyanin.

GC-MS analysis of Methanolic extract of *Curcuma angustifolia*, *Curcuma decipiens*, and *Curcuma longa*

Methanolic extracts of selected species were further tested by gas chromatography – mass spectrometry for detailed screening of metabolites.

Analysis of these samples were performed on instrument shimadzu qp2010 ultra.

Discription of GC-MS parameter

Column Details- Rxi: 5ms, length: 30.0 m, thickness: 0.25 μ m, Diameter: 0.25 mm.

Gas chromatography Details- Column oven temperature : 40 °C, injection temperature : 310 °C, injection mode : split, split ratio : 20:1, flow control mode: linear velocity 32.3 cm/sec, pressure: 33.8 kPa, purge flow: 3.0 ml/min, column flow : 0.80 ml/min, oven ramp : 40 °C holds for 2.0 min: 10 °C/min to 100 °C holds for 3.0 min and 10 °C/min to 320 °C holds for 2.0 min, total run time 35.00 min.

Mass spectrometry Details- Ion source temperature: 200 °C, interface temperature: 330 °C, solvent cut time: 0.0 min, detector voltage: 0.7 kV, acquisition mode: Scan mode, scan speed: 3333, event time: 0.30 sec, starting m/z: 40 to 999 m/z.

Result

Result of comparative preliminary phytochemical screening

Table 1: Preliminary phytochemical screening of *Curcuma angustifolia*, *Curcuma decipiens* and *Curcuma longa*

Test	<i>Curcuma angustifolia</i>			<i>Curcuma decipiens</i>			<i>Curcuma longa</i>		
	Methanol	Ethanol	Acetone	Methanol	Ethanol	Acetone	Methanol	Ethanol	Acetone
Flavonoid	-	-	-	-	-	-	+	+	+
NH ₄ OH test	-	-	-	-	-	-	+	+	+
Z _n dust test	+	-	+	+	+	-	+	+	-
Carbohydrate	+	+	-	+	+	-	+	+	+
Fehling test	+	+	+	+	+	+	+	+	+
Molisch test	+	+	-	-	+	+	+	-	+
Iodine test	-	-	+	-	+	+	-	+	-
Protein	-	+	+	-	+	+	-	+	-
Xanthoprotic test	-	-	-	-	-	-	+	+	+
Biuret test	-	-	-	-	-	-	+	+	+
Alkaloid	+	+	+	+	+	+	+	+	+
Steroid	+	+	+	+	+	+	-	+	+
Tanin	+	+	+	+	+	+	+	+	+
Saponin	-	-	+	+	+	-	-	-	+
Fatty acids & oil	+	+	+	+	+	+	+	+	+
Amino acids	+	+	-	+	+	-	+	+	-
Ninhydrin test	+	+	-	+	+	-	+	+	-
Anthocyanin	+	-	-	-	-	-	+	+	+

Result of GC-MS analysis**1. *Curcuma angustifolia***

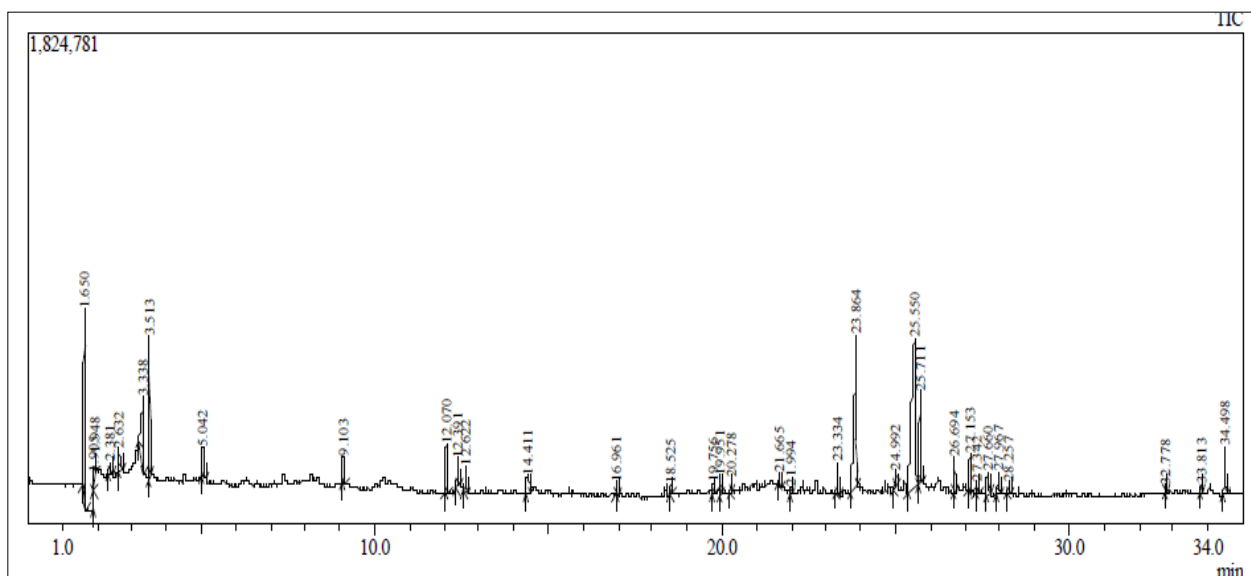
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GC-MS Chromatogram of methanolic extract of *Curcuma angustifolia***Table 2:** Phytochemical compounds of methanolic extract of *Curcuma angustifolia*

Peak#	RT	Peak Area%	Peak Height%	Molecular Weight	Molecular Formula	Name of compound
1	1.650	12.31	13.52	44	N ₂ O	Nitrous oxide
2	1.905	1.48	1.99	40	C ₃ H ₄	Allene
3	1.948	1.11	2.25	46	C ₂ H ₆ O	Ethanol
4	2.381	0.75	0.88	86	C ₄ H ₆ O ₂	Acetic acid ethenyl ester
5	2.632	1.47	1.64	99	CH ₂ O ₂	Formic acid
6	3.338	6.01	5.19	60	C ₃ H ₄ O ₂	Acetic acid
7	3.513	4.85	10.00	74	C ₃ H ₆ O ₂	2-Propanone, 1-hydroxy-
8	5.042	1.31	2.07	102	C ₄ H ₆ O ₃	Propanoic acid, 2-oxo-, methyl ester
9	9.103	1.04	1.89	154	C ₁₀ H ₁₈ O	Eucalyptol (Cineole)
10	12.070	2.53	3.47	152	C ₁₀ H ₁₆ O	(+)-2-Bornanone (camphor)
11	12.391	1.42	2.01	154	C ₁₀ H ₁₈ O	Isoborneol
12	12.622	1.38	1.91	154	C ₁₀ H ₁₈ O	(-)-Borneol
13	14.411	2.31	1.33	126	C ₆ H ₆ O ₃	5-Hydroxymethylfurfural
14	16.961	0.44	0.88	204	C ₁₅ H ₂₄	Cyclohexane, 1-ethenyl-1-methyl-2,4-bis(1-methylethenyl)
15	18.525	0.50	0.73	216	C ₁₅ H ₂₀ O	Benzofuran, 6-ethenyl-4,5,6,7-tetrahydro-3-, (Curzerene)
16	19.756	0.66	0.79	220	C ₁₅ H ₂₄ O	Caryophyllene oxide
17	19.951	0.81	1.33	218	C ₁₅ H ₂₂ O	Germacron
18	20.278	0.50	1.03	220	C ₁₅ H ₂₄ O	1H-Cycloprop[e]azulen-7-ol,
19	21.665	0.59	0.89	228	C ₁₄ H ₂₈ O ₂	Tetradecanoic acid
20	21.994	0.37	0.54	220	C ₁₅ H ₂₄ O	(-)-Spathulenol
21	23.334	1.10	2.28	270	C ₁₇ H ₃₄ O ₂	Hexadecanoic acid, methyl ester
22	23.864	16.67	10.75	256	C ₁₆ H ₃₂ O ₂	n-Hexadecanoic acid

23	24.992	0.58	1.27	294	C ₁₉ H ₃₄ O ₂	8,11-Octadecadienoic acid, methyl ester
24	25.550	23.45	10.45	282	C ₁₈ H ₃₄ O ₂	Oleic Acid
25	25.711	5.55	6.32	284	C ₁₈ H ₃₆ O ₂	Octadecanoic acid
26	26.694	1.44	2.48	312	C ₂₁ H ₄₄ O	1-Heneicosanol
27	27.153	2.03	2.75	246	C ₁₅ H ₁₈ O ₃	4,7-Methanofuro[3,2-c]oxacycloundecin-6(4H)-one
28	27.153	0.57	0.81	312	C ₂₀ H ₄₀ O ₂	Eicosanoic acid
29	27.660	1.19	1.59	228	C ₁₄ H ₁₂ O ₃	Trioxsalen
30	27.967	1.49	1.65	308	C ₁₇ H ₂₄ O ₅	Naphtho[2,3-b]furan-2(3H)-one
31	28.257	0.73	0.83	308	C ₁₇ H ₂₄ O ₅	Naphtho[2,3-b]furan-2(3H)-one
32	32.778	0.31	0.54	384	C ₂₇ H ₄₄ O	Cholesta-4,6-dien-3-ol, (3.beta.)-
33	33.813	0.41	0.56	400	C ₂₈ H ₄₈ O	:Campesterol
34	34.498	2.64	3.37	414	C ₂₉ H ₅₀ O	gamma.-Sitosterol

2. Curcuma decipiens

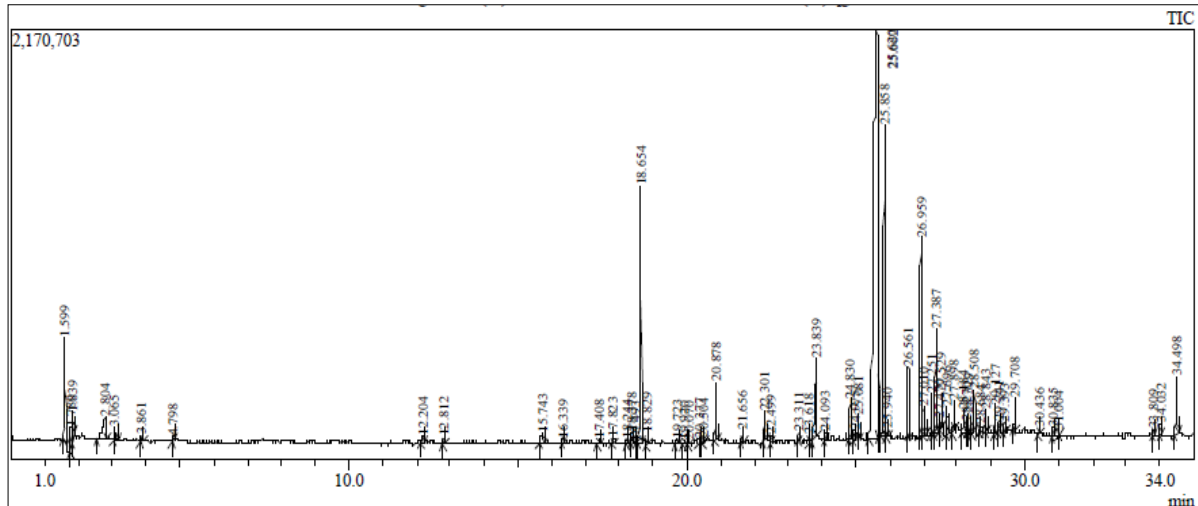
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GC-MS Chromatogram of methanolic extract of *Curcuma decipiens***Table 3:** Phytochemical compounds of methanolic extract of *Curcuma decipiens*

Peak#	RT	Peak Area%	Peak Height%	Molecular Weight	Molecular Formula	Name of compound
1	1.599	3.20	3.71	44	N ₂ O	Nitrous oxide
2	1.780	0.60	0.76	40	C ₃ H ₄	Allene
3	1.839	0.54	0.99	46	C ₂ H ₆ O	Ethanol
4	2.804	1.55	0.28	60	C ₂ H ₄ O ₂	Acetic acid
5	3.065	0.41	0.43	74	C ₃ H ₆ O ₂	2-Propanone, 1-hydroxy
6	3.861	0.23	0.24	92	C ₃ H ₈ O ₃	Glycerin
7	4.798	0.11	0.15	102	C ₄ H ₆ O ₃	Propanoic acid, 2-oxo-, methyl ester
8	12.204	0.32	0.25	144	C ₆ H ₈ O ₄	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy
9	12.812	0.16	0.27	154	C ₁₀ H ₁₈ O	Terpinen-4-ol
10	15.743	0.56	0.38	170	C ₁₀ H ₁₈ O ₂	2-Oxabicyclo[2.2.2]octan-6-ol, 1,3,3-trimethyl (2-hydroxy-1,8 cineole)
11	16.339	0.10	0.15	192	C ₁₄ H ₂₄	1,8-Nonadiene, 2,7-dimethyl-5-(1-methylethe
12	17.408	0.13	0.20	204	C ₁₅ H ₂₄	Caryophyllene
13	17.823	0.11	0.23	204	C ₁₅ H ₂₄	cis-.beta.-Farnesene
14	18.244	0.09	0.18	202	C ₁₅ H ₂₂	alpha curcumene
15	18.378	0.19	0.39	518	C ₁₄ H ₄₂ O ₇ Si ₇	Cycloheptasiloxane, tetradecamethyl-
16	18.493	0.03	0.08	236	C ₁₆ H ₂₈ O	4-Hexen-1-ol, 6-(2,6,6-trimethyl-1-cyclohexe
17	18.654	6.95	0.17	204	C ₁₅ H ₂₄	beta.-Bisabolene
18	18.829	0.21	0.42	204	C ₁₅ H ₂₄	beta.-Sesquiphellandrene
19	19.723	0.26	0.21	220	C ₁₅ H ₂₄ O	Caryophyllene oxide
20	19.920	0.12	0.15	220	C ₁₅ H ₂₄ O	Lanceol, cis
21	20.070	0.10	0.14	220	C ₁₅ H ₂₄ O	Farnesene epoxide, E-
22	20.377	0.16	0.15	208	C ₁₄ H ₂₄ O	2-Methyl-4-(2,6,6-trimethylcyclohex-1-enyl)
23	20.504	0.10	0.19	592	C ₁₆ H ₄₈ O ₈ Si ₈	Cyclooctasiloxane, hexadecamethyl-
24	20.878	1.23	2.07	222	C ₁₅ H ₂₆ O	.alpha.-Bisabolol
25	21.656	0.29	0.43	206	C ₁₄ H ₂₂ O	2-Butenal, 2-methyl-4-(2,6,6-trimethyl-1-cyclohexen
26	22.301	0.68	1.04	236	C ₁₆ H ₂₈ O	4-Hexen-1-ol, 6-(2,6,6-trimethyl-1-cyclohexenyl
27	22.499	0.15	0.27	186	C ₁₁ H ₂₂ O ₂	Octanal, 7-methoxy-3,7-dimethyl
28	23.311	0.15	0.35	270	C ₁₇ H ₃₄ O ₂	Hexadecanoic acid, methyl ester
29	23.618	0.15	0.28	190	C ₁₄ H ₂₂	1,3-Cyclopentadiene, 2,3,4,5-tetramethyl-1-(4-pentenyl

30	23.839	3.66	2.82	256	C ₁₆ H ₃₂ O ₂	n-Hexadecanoic acid
31	24.093	0.24	0.35	232	C ₁₅ H ₂₀ O ₂	Germacra-1(10),4,11(13)-trien-12-oic acid, 6.alpha.-hydroxy-,gamma.-lactone,
32	24.830	0.77	1.40	284	C ₂₀ H ₂₈ O	Retinal, 9-cis-
33	24.975	0.28	0.36	236	C ₁₆ H ₂₈ O	4-Hexen-1-ol, 6-(2,6,6-trimethyl-1-cyclohexenyl)-4-methyl-,
34	25.081	0.38	0.73	444	C ₃₀ H ₅₂ O ₂	Tricyclo[20.8.0.0(7,16)]triacontane, 1(22),7(16)-diepoxy
35	25.620	32.81	14.56	206	C ₁₃ H ₁₈ O ₂	4,4-Dimethyl-2,4,5,6-tetrahydro-1H-inden-2-yl)acetic acid
36	25.662	8.51	14.22	334	C ₁₈ H ₃₉ O ₃ P	Ethylphosphonic acid, di(2-ethylhexyl) ester
37	25.858	9.48	11.11	206	C ₉ H ₁₉ O ₃ P	Cycloheptyl ethyl methylphosphonate
38	25.940	0.19	0.28	318	C ₂₀ H ₃₀ O ₃	Hydroxydehydrostevic acid
39	26.561	1.45	2.48	206	C ₁₅ H ₂₆	(+)-(Z)-Longipinane
40	26.959	8.56	7.08	288	C ₂₁ H ₃₆	Trispiro[4.2.4.2.4.2.]heneicosane
41	27.010	0.60	1.04	206	C ₁₄ H ₂₂ O	2,3,3-Trimethyl-2-(3-methyl-buta-1,3-dienyl)-cyclohexanone
42	27.251	0.74	1.43	194	C ₁₃ H ₂₂ O	5,5,8a-Trimethyldecalin-1-one
43	27.387	3.60	3.69	194	C ₁₃ H ₂₂ O	:5,5,8a-Trimethyldecalin-1-one
44	27.470	0.24	0.44	194	C ₁₃ H ₂₂ O	5,5,8a-Trimethyldecalin-1-one
45	27.529	0.62	1.27	368	C ₁₈ H ₂₅ F ₅ O ₂	(-)-Isolongifolol, pentafluoropropionate
46	27.696	0.92	0.99	334	C ₂₁ H ₃₄ O ₃	Methyl dihydroisosteviol
47	27.898	0.65	1.08	194	C ₁₁ H ₁₄ O ₃	2H-Inden-2-one, 1,4,5,7a-tetrahydro-6,7-bis(hydroxymethyl)
48	28.184	0.85	0.80	194	C ₁₃ H ₂₂ O	5,5,8a-Trimethyldecalin-1-one
49	28.270	0.29	0.38	400	C ₂₈ H ₄₈ O	Cholest-14-en-3-ol, 4-methyl-, (3. beta., 4.alpha., 5.alpha.)
50	28.329	0.39	0.65	226	C ₁₃ H ₂₂ O ₃	3H-3,10a-Methano-1,2-benzodioxocin-3-ol, octahydro-7,7-dimethyl-,
51	28.508	0.95	1.47	272	C ₂₀ H ₃₂	alpha kaurene
52	28.684	0.11	0.26	374	C ₂₄ H ₃₈ O ₃	17-Oxo-6-pentyl-4-nor-3,5-seco-5-androsten-3-oic acid, methyl ester
53	28.843	0.48	0.77	332	C ₂₀ H ₂₈ O ₄	3-Keto-isosteviol
54	29.127	0.62	1.04	236	C ₁₆ H ₂₈ O	4-Hexen-1-ol, 6-(2,6,6-trimethyl-1-cyclohexenyl)-4-methyl-, (E)-
55	29.241	0.31	0.32	234	C ₁₅ H ₂₂ O ₂	2-(4a,8-Dimethyl-6-oxo-1,2,3,4,4a,5,6,8a-octahydro-naphthalen-2-yl)-propionaldehyde
56	29.393	0.29	0.32	222	C ₁₅ H ₂₆ O	1-Naphthalenemethanol, 1,4,4a,5,6,7,8,8a-octahydro-2,5,5,8a-tetramethyl-
57	29.708	0.72	1.16	248	C ₁₂ H ₁₈ Cl ₂ O	Bicyclo[8.2.0]dodecan-11-one, 12,12-dichloro-, (1R*,10S*)-
58	30.436	0.19	0.25	208	C ₁₄ H ₂₄ O	2-Methyl-4-(2,6,6-trimethylcyclohex-1-enyl)but-2-en-1-ol
59	30.835	0.17	0.29	194	C ₁₁ H ₁₈ N ₂ O	1-Pyridineethanamine, beta.-(2-furanyl)hexahydro
60	31.004	0.08	0.16	220	C ₁₄ H ₂₀ O ₂	2-Cyclohexen-1-one, 4-hydroxy-3,5,5-trimethyl-4-(3-methyl-1,3-butadienyl)-, [S-(E)]-
61	33.809	0.13	0.22	400	C ₂₈ H ₄₈ O	Campesterol
62	34.032	0.25	0.42	412	C ₂₉ H ₄₈ O	Stigmasterol
63	34.498	1.54	2.03	414	C ₂₉ H ₅₀ O	.gamma.-Sitosterol

3. Curcuma longa

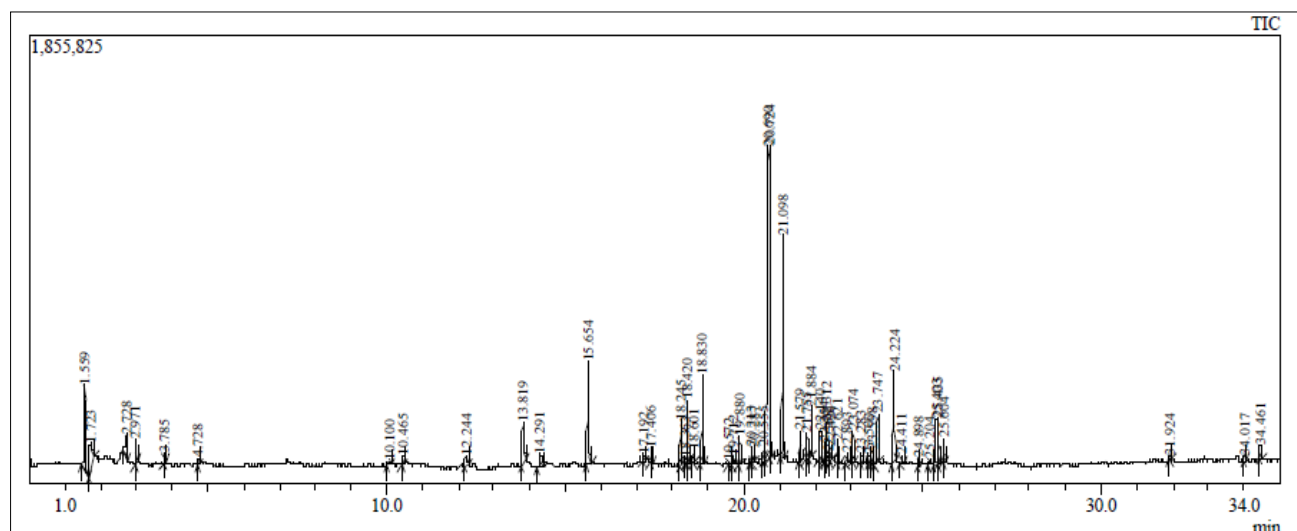
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GC-MS Chromatogram of methanolic extract of *Curcuma longa*

Table 4: Phytochemical compounds of methanolic extract of *Curcuma longa*

Peak#	RT	Peak Area%	Peak Height%	Molecular Weight	Molecular Formula	Name of compound
1	1.559	3.13	3.93	44	CO ₂	Carbon dioxide
2	1.723	2.63	1.32	40	C ₃ H ₄	Allene
3	2.728	1.40	1.13	60	C ₂ H ₄ O ₂	Acetic acid
4	2.971	0.90	1.14	74	C ₃ H ₆ O ₂	2-Propanone, 1-hydroxy-

5	3.785	0.46	0.41	92	C ₃ H ₈ O ₅	Glycerin
6	4.728	0.38	0.32	102	C ₄ H ₆ O ₃	Propanoic acid, 2-oxo-, methyl ester
7	10.100	0.43	0.21	128	C ₆ H ₈ O ₃	2,5-Dimethyl-4-hydroxy-3(2H)-furanone
8	10.465	0.59	0.42	124	C ₇ H ₈ O ₂	Phenol, 2-methoxy
9	12.244	0.81	0.45	144	C ₆ H ₈ O ₄	4H-Pyran-4-one,2,3-dihydro-3,5-dihydroxy-6-methyl-
10	13.819	2.87	1.97	120	C ₈ H ₈ O	Benzofuran, 2,3-dihydro(Coumaran)
11	14.291	1.61	0.65	126	C ₆ H ₆ O ₃	5-Hydroxymethylfurfural
12	15.654	5.11	4.76	150	C ₉ H ₁₀ O ₂	2-Methoxy-4-vinylphenol
13	17.192	0.59	0.36	152	C ₈ H ₈ O ₃	Vanillin
14	17.406	0.48	0.69	204	C ₁₅ H ₂₄	Caryophyllene
15	18.245	1.44	2.10	202	C ₁₅ H ₂₂	alpha- curcumene
16	18.363	0.17	0.26	518	C ₁₄ H ₄₂ O ₇ Si ₇	Cycloheptasiloxane, tetradecamethyl-
17	18.420	2.03	2.99	204	C ₁₅ H ₂₄	Zingiberene
18	18.601	0.40	0.59	204	C ₁₅ H ₂₄	beta -Bisabolene
19	18.830	3.06	4.12	204	C ₁₅ H ₂₄	beta.-Sesquiphellandrene
20	19.572	0.18	0.19	238	C ₁₈ H ₂₂	Dicumene
21	19.712	0.46	0.41	220	C ₁₅ H ₂₄ O	Caryophyllene oxide
22	19.880	1.65	1.36	162	C ₁₂ H ₁₈	Benzene, 1,4-dimethyl-2-(2-methylpropyl)
23	20.213	0.53	0.77	222	C ₁₅ H ₂₆ O	7-epi-cis-sesquisabinene hydrate
24	20.287	0.67	0.66	218	C ₁₅ H ₂₂ O	Curlone
25	20.555	0.63	0.62	218	C ₁₅ H ₂₂ O	Tumerone
26	20.690	18.41	14.52	216	C ₁₅ H ₂₀ O	Ar-tumerone
27	20.724	9.52	14.58	218	C ₁₅ H ₂₂ O	Tumerone
28	21.098	10.17	10.36	218	C ₁₅ H ₂₂ O	Curlone
29	21.579	1.09	1.45	290	C ₁₉ H ₃₀ O ₂	Cyclopentanecarboxylic acid, 3-isopropylidene-, bornyl ester
30	21.755	1.83	1.20	162	C ₆ H ₁₄ O ₃ Si	Trimethylsilyl 2-methoxyacetate
31	21.884	1.70	2.32	218	C ₁₅ H ₂₂ O	Germacon
32	22.140	1.23	1.44	350	C ₁₄ H ₁₇ F ₇ O ₂	Isoborneol, heptafluorobutyrate (ester)
33	22.254	0.78	0.93	220	C ₁₅ H ₂₄ O	1-Isopropyl-4,8-dimethylspiro[4.5]dec-8-en-7-one
34	22.312	1.52	1.81	250	C ₁₆ H ₂₆ O ₂	Methyl 5,5,8a-trimethyl-2-methylenedecahydro
35	22.368	0.16	0.24	172	C ₁₀ H ₁₇ Cl	Bornyl chloride(Turpentine camphor)
36	22.424	0.54	0.60	192	C ₁₁ H ₁₂ O ₃	3-Buten-2-one, 4-(4-hydroxy-3-methoxyphenyl)- (Dehydrogingiberon)
37	22.621	0.98	1.02	208	C ₁₁ H ₁₂ O ₄	2-Propenoic acid, 3-(4-hydroxy-3-methoxyphenyl)-, methyl ester (Cinnamic acid)
38	22.893	0.48	0.46	140	C ₉ H ₁₆ O	2-Methyl-4-octenal
39	23.074	1.03	1.48	246	C ₁₆ H ₂₂ O ₂	3-Phenylpropyl cyclohexanecarboxylate
40	23.283	0.67	0.49	220	C ₁₅ H ₂₄ O	6Z-2,5,5,10-Tetramethyl-undeca-2,6,9-trien-8-one (1R,4S)-1,7,7-Trimethylbicyclo[2.2.1]heptan-2-yl (E)-2- methylbut-2-enoate
41	23.500	0.40	0.36	236	C ₁₅ H ₂₄ O ₂	(E)-2-Isopropyl-5-methylphenyl 2-methylbut-2-enoate
42	23.598	0.52	0.67	232	C ₁₅ H ₂₀ O ₂	n-Hexadecanoic acid (Palmitic acid)
43	23.747	2.77	2.30	256	C ₁₆ H ₃₂ O ₂	2-Butenoic acid, 2-methyl-, 2-(acetyloxy)-1,
44	24.224	5.50	4.22	490	C ₂₇ H ₃₈ O ₈	2-Butenoic acid, 2-methyl-, 2-(acetyloxy)-1,
45	24.411	0.40	0.43	490	C ₂₇ H ₃₈ O ₈	2-Butenoic acid, 2-methyl-, 2-(acetyloxy)-1,
46	24.898	0.30	0.40	126	C ₈ H ₁₄ O	2-Hepten-4-one, 2-methyl
47	25.204	0.45	0.31	154	C ₁₀ H ₁₈ O	3-Decen-5-one
48	25.403	2.89	2.13	280	C ₁₈ H ₃₂ O ₂	9,12-Octadecadienoic acid (Z,Z) (Linolic acid)
49	25.435	2.12	2.20	282	C ₁₈ H ₃₄ O ₂	9-Octadecenoic acid
50	25.604	0.91	1.13	284	C ₁₈ H ₃₆ O ₂	Octadecanoic acid/ Stearic acid (Hystrene S-97)
51	31.924	0.19	0.21	452	C ₃₁ H ₃₆ N ₂ O	Ezloptant, dehydro-
52	34.017	0.22	0.24	412	C ₂₉ H ₄₈ O	Stigmasterol
53	34.461	0.65	0.66	414	C ₂₉ H ₅₀ O	.gamma.-Sitosterol

Discussion

After analysis of GC-MS chromatograms of methanolic extract of *Curcuma angustifolia*, *Curcuma decipiens*, and *Curcuma longa*, results were compared with each other. *Curcuma angustifolia* showed 34 peaks representing the presence of 34 phytochemical compounds with retention time and percent different areas. Likewise *Curcuma decipiens* showed 63 peaks of phytochemicals and *Curcuma longa* showed 53 peaks of phytochemicals with retention time and percent areas. When mass spectra of these compounds were compared with NIST11s library, then certain phytochemical components identified by retention time, molecular weight, molecular formula, and percent area. Compounds have structural similarity with NIST11s library is ≥ 98 SI. (Similarity index).

In *Curcuma angustifolia* there were some diverse bioactive compounds those covers more area percent include oleic acid (23.5%), n-hexadecanoic acid (16.67%), nitrous oxide(12.31%), acetic acid (6.01%), octadecanoic acid (5.55%), 2-propanone 1-hydroxy (4.85%), gama sitosterol

(2.64%), (+)-2-bornanone (2.53%), 5-hydroxymethylfurfural(2.31%), 4,7-methanofuro[3,2-c]oxacycloundecin-6 (2.03%), naphtho[2,3-b]furan-2(3H)-one, 4-(acetyloxy (1.49%), 1-heneicosanol (1.44%), isoborneol (1.44%), borneol (1.38%), propanoic acid, 2-oxo-, methyl ester (1.31%), eucalyptol (1.04%).

In case of *Curcuma decipiens major* metabolites were include 4, 4-Dimethyl-2, 4, 5, 6-tetrahydro-1H-inden-2-yl) acetic acid (32.81%), cycloheptyl ethyl methylphosphonate (9.48%), Trispiro [4.2.4.2.4.2.] heneicosane (8.56%), ethylphosphonic acid, di(2-ethylhexyl) ester (8.51%), beta.-bisabolene (6.95%), n-hexadecanoic acid (3.66%), 5,5,8a-trimethyldecalin-1-one (3.60%), acetic acid (1.55%), (+)-(Z)-longipinane (1.45%), alpha.-bisabolol (1.23%), methyl dihydroisosteviol (0.96%), kaur-16-ene (0.95%).

In *Curcuma longa*, higher percent area phytochemicals were include ar-tumerone (18.41%), curlone (10.17%), tumerone (9.52%), 2-butenic acid, 2-methyl-, 2-(acetyloxy)-1,1a (5.50%), 2-methoxy-4-vinylphenol (5.11%), carbon dioxide (3.13%), cyclohexene, 3-(1,5-dimethyl-4-hexenyl)-6-

methylene (3.06%), 9,12-octadecadienoic acid (Z,Z)-(2.89%), Benzofuran, 2,3-dihydro- (2.87%), n-Hexadecanoic acid (2.77%), allene (2.63%), 1,3-cyclohexadiene, 5-(1,5-dimethyl-4-hexenyl (2.03%), trimethylsilyl 2-methoxyacetate (1.83%), 3,7-Cyclodecadien-1-one, 3,7-dimethyl-10- (1.70%), benzene, 1,4-dimethyl-2-(2-methylpropyl)- (1.65%), 5-

hydroxymethylfurfural (1.61%), alpha- curcumene (1.44%), Acetic acid (1.40%).

Comparison of phytoconstituents of *Curcuma longa* with phytoconstituents of wild species *Curcuma angustifolia* and *Curcuma decipiens* and their biological activity is given below.

Table 5: Comparative phytochemical analysis and their biological activity

S. No	Name of compound	Curcuma longa	Curcuma angustifolia	Curcum decipiens	Biological activity
1.	Allene	+	+	+	Not reported
2.	Acetic acid	+	+	+	anti-bacterial; anti-otitic; anti-salmonella; anti-vaginitic; expectorant; acidulant; fungicide
3.	2-Propanone,1-hydroxy	+	+	+	preservative
4.	Glycerine	+	-	+	antibacterial
5.	Propanoic acid, 2-oxo-, methyl ester	+	+	+	flavor, fungicide, irritant, perfumery, pesticide
6.	4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy	+	-	+	anti-microbial, anti-inflammatory, anti-proliferative anti-oxidant, automatic nerve activity
7.	5-Hydroxymethylfurfural	+	+	-	antioxidant, cancer chemoprevention.
8.	Caryophyllene	+	-	+	allergenic, analgesic, antiacne, antiasthematic, antibacterial, anticariogenic, antidermatitic, antidemic, anti-inflammatory, antileishmanic, antiproliferant, antistaphylococci, antistreptococci, antitumor, candidicide, flavor, gastroprotective, larvicide mosquitocide, pesticide, sedative,
9.	Alpha curcumene	+	-	+	Anti-inflammatory antitumor hypotriglyceridemic
10.	beta-Bisabolene	+	-	+	abortifacient, antirhinoviral antiulcer, antiviral, perfumery, stomachic
11.	beta.-Sesquiphellandrene	+	-	+	antirhinoviral, antiulcer, anticancer, expectorant, pesticide, stomachic
12.	Caryophyllene oxide	+	+	+	antimicrobial, anti-inflammatory, antioxidant, antidermatitis, fragrances, flavors
13.	Germacon	+	+	-	Analgesic antiedemic anti-inflammatory antistress antitussive antiulcer hypothermic
14.	n-Hexadecanoic acid	+	+	+	antioxidant, anti-inflammatory
15.	Octadecanoic acid	+	+	-	antibacterial, antiviral, antioxidant
16.	Stigmasterol	+	-	+	antihepatotoxic, anti-inflammatory, antinociceptive, antiophidic, antioxidant, antiviral, cancer-preventive, estrogenic, hypocholesterolemic, ovulant, sedative
17.	gamma-Sitosterol	+	+	+	antidiabetic, antibacterial, anti-angeogenic, anticancer, anti-inflammatory, antidiarrhoeal, antiviral

Sixteen Compounds out of seventeen mentioned in table 5, exhibit biological activity. Compounds such as acetic acid shows antibacterial activity against the micro-organism *Pseudomonas aeruginosa* [9, 10], 2-propanone,1-hydroxy used as preservative [9, 11], glycerine has antibacterial activity [12], propanoic acid, 2-oxo-, methyl ester used as pesticide [9], 4H-Pyran-4-one, 2,3-dihydro-3,5-dihydroxy used as antimicrobial [9, 13], 5-Hydroxymethylfurfural shows antioxidant activity [14], caryophyllene shows anti-allergic and multiple activities [15], alpha curcumene exhibits anti-inflammatory and antitumor activities [15], beta bisabolene shows abortifacient and multiple activities [15], beta sesquiphellandrene shows antirhinoviral and multiple activity [15], caryophyllene oxide also shows antimicrobial and multiple activities [16, 17], germacon used in analgesic and multiple activities [15], n-hexadecanoic acid used in antioxidant and anti-inflammatory activity [18], octadecanoic acid shows antibacterial, antiviral activity [19], stigmasterol shows antihepatotoxic and multiple activities [15], gamma-sitosterol shows antidiabetic and multiple activities [17, 20].

Conclusion

In this investigation pharmacognostical parameters such as preliminary phytochemical screening and GC-MS profiling of *Curcuma longa*, *Curcuma angustifolia*, and *Curcuma decipiens* were carried out. However *Curcuma longa* has been studied previously by many researchers. Those investigations illustrate that it is most common cultivated species which is rich in many phytoconstituents that exhibit several medicinal use and biological activity, but there was necessity to compare the phytoconstituents of *Curcuma longa* with phytoconstituents of other wild species of genus *Curcuma*. The results obtained in this study indicate that 34 phytochemicals were found in *Curcuma angustifolia*, 63

phytochemicals were found in *Curcuma decipiens* and 53 phytochemicals were found in *Curcuma longa*. The 10 phytochemical compounds of *Curcuma longa* were identical with *Curcuma angustifolia* and 14 with *Curcuma decipiens*, while other phytochemicals of these wild species were different from *Curcuma longa* and show evidence of significant biological activity. Further studies of the present investigation may suggest for the isolation of bioactive constituents and biological assay methods from selected wild species for the standard drug preparations.

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