Pharmacogn J. 2017; 9(6)Suppl: s28-s34
A Multifaceted Journal in the field of Natural Products and Pharmacognosy
www.phcogj.com | www.journalonweb.com/pj | www.phcog.net

Original Article

Distribution, Ethnobotany, Pharmacognosy and Phytoconstituents of *Coptis teeta* Wall.: A Highly Valued and Threatened Medicinal Plant of Eastern Himalayas

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ABSTRACT

Objectives: To study the distribution, traditional knowledge, dose and preparations, phytoconstituents, pharmacognostic characters and to correlate phytoconstituents and the claimed health benefits among the tribal people of Arunachal Pradesh if any, the present study was carried out. **Methods:** Field survey and interview were used for ethnobotany and protocol given in Shah and Seth (2010), Kakote *et. al.*,(2012) and Wallis (2011) was followed to study pharmacognostic studies while GC-MS was used in the studies of phytoconstituents. **Results:** This study reports numbers of volatile and non-volatile compounds from the rhizome of *Coptis teeta* with high percentage of berberine alkaloids. The study also reports alkaloid deposition at parenchymatous tissues and vascular tissues of rhizome. Phytoconstituents presents in the ethanolic extract of *C.teeta* cold be related to health problems and phytoconstituents as claimed by tribal people of Arunachal Pradesh. **Conclusion:** Locally called Mishmi teeta in Arunachal Pradesh, *Coptis teeta* Wall. is a well-known medicinal plant used among Mishmi and other tribes of Arunachal Pradesh for health problems like loose motion, stomach pain, diarrhoea and malaria. This endemic and threatened medicinal plant contains numbers of biologically active compounds and need *in-situ* as well as *ex-situ* conservation.

Key words: Mishmi tribe, Medicinal plant, Threatened, Phytoconstituents, Eastern Himalayas, Berberine.

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History

- Submission Date: 18-10-2017;
- Review completed: 08-11-2017;
- Accepted Date: 20-11-2017

DOI : 10.5530/pj.2017.6s.154

Article Available online
http://www.phcogj.com/v9/i6s

INTRODUCTION

Nature has endowed plant kingdom to the blue planet and plant is one among pre-requisite life supporting system. Plant provides almost everything that man needed and the environment around has been influencing the man's culture since antiquity to make use of available resources around him. Therefore, Man must have learnt use of medicinal plant subject to the hungry, accident, injury, bite of insects or serpent or death of fellow men and made man's instinct by luck or observation to get help from available surroundings [1] thus the importance of plants and animals was realized by the early men since the dawn of time. They started to use fruits, leaves and tubers for food and medicine. Such experiences became a knowledge system and assimilated as an integral part of man culture that passed down generation after generation and climaxed into *traditional knowledge* system.[2] Therefore, Knowledge on the therapeutic and nutritional properties of medicinal herbs and food plants predated recorded history.[3] Likewise, there are several historical indications that claim the ancient use of natural remedies to resolve primary health problems in different part of the world.[1,4,5] In fact, traditional medicine practice is as old as mountains and hills in a tribal society where primary health care is totally based on traditional medicines.[2] With the emerging worldwide interest in adopting and studying traditional systems and exploring their potential based on different health care systems, the evaluation of rich heritage of traditional medicine is essential.[6]

Moreover, in more recent past, the use of plants as medicines has involved the isolation of active compounds, beginning with the isolation of morphine from the opium in the early 19th century.[7] Drug analysis from medicinal plants led to the isolation of important drugs like cocaine, codeine, digitoxin, and quinine.[8]

As cited above, Indigenous people of Arunachal Pradesh also has predated knowledge on medicinal plants and their uses. But no written records is exist in this part of territory however still exists in form of *traditional knowledge* and need to record before it is lost forever. It was the Britishers, who explored flora including their uses among aborigines of Arunachal Pradesh in late Nineteenth Century. Mishmi teeta is one among the herbal treasure grows in Arunachal Pradesh. *Coptis teeta* has been reported only from Indian Territory of Arunachal Pradesh and Yunnan province of China. This herb grows at the lower elevation limit of 1700 mts and upper elevation limit of 2800 mts. This herb is enlisted in Red List Category as endangered (A2cd ver 3.1) (IUCN Red List of threaneted species). The combined effects of unregulated collection and degradation of forest over the

Cite this article: Payum T. Distribution, Ethnobotany, Pharmacognosy and Phytoconstituents of *Coptis teeta* Wall.: A Highly Valued and Threatened Medicinal Plant of Eastern Himalayas. Pharmacog J. 2017;9(6)Suppl:s28-s34.

last decade years has resulted in decline of the population of over 60%. Therefore, the species has been assessed as Endangered. About 90% *C. teeta* population is in India, therefore considered representative of that of the global population.[9]

This herb was recorded as early as 1825 by R. Wilcox and Captain Bedford followed by Griffith in 1836 from Mishmi Hills of Indian flora. Mishmis and other tribes of Arunachal Pradesh used *C. teeta* to treat malaria, stomach ache and dysentery. The current rate of *Coptis teeta* is about Rs.2000/kg, it is found to be adulterated with the rootstock of *Geranium wallichianum D*. Don ex Sweet, root of *Thalictrum filiolosum* DC, roots of *Swertia* genus resembles the of rhizome of *C. teeta.*[10]

In the word of Philipson,[11] herbal medicines are, of course, used for their reputed beneficial effects, however, scientific studies for validation are also important and Pharmacognosy is one of the basic methods to characterize and validate the drugs of natural origin to give correct and authentic identity. Plants provide a great challenge in metabolomics due to the rich chemical diversity of metabolites that they possess across a huge range of concentrations; estimates of 100000-200000 metabolites have been made for the plant kingdom,[12] And Gas Chromatography–Mass Spectrometry (GC-MS) is a hyphenated analytical technique that combines the separation properties of gas-liquid chromatography with the detection feature of mass spectrometry to identify different substances within a test sample.[13] Gas chromatography-mass spectrometry (GC-MS) is the most commonly used technique for the analysis of liposoluble constituents, especially volatile/semi-volatile compounds, and their metabolites in biological fluids due to its high resolution, selectivity and sensitivity.[14] Hence, the present study has been taken up to study phytocompounds present in the Mishmi teeta by using GC-MS and pharmacognostic parameters of *Coptis teeta.*

Figure 1: *Coptis teeta*.

Figure 2: Dried rhizome.

MATERIALS AND METHODS

Plant material

Coptis teeta is a stem less herbaceous plant of *Ranunculceae* family, much used in Bengal and elsewhere as recorded by Kanjilal,[15] the roots are gathered towards the end of the rainy season and are carried in tiny little wicker baskets to Sadiya where dealers from other provinces go to buy them.[13] This herb is perennial herb, rhizomatous with densely fibrous roots, petiole is long, leaves are *ovate – lanceolate,* flowers are whitish-yellow bloom in the month of Feb'- April and rhizome is golden in colour and bitter in taste Figure 1 and 2.

Pharmacognostic studies

For pharmacognostic works, fresh rhizome was collected from Hawaii forest by Miss Amina Miso of BSc- Botany major Vth semester student of J.N.College, Pasighat. Characterization of pharmacognostic parameters were carried out by following methods given in Shah and Seth.[16] Kakote *et al.*[17] Wallis.[18]

Preparation of extract

The dry rhizome of *Coptis teeta* was collected from Anini by Y. Jamoh Lego (a local healer) of Arunachal Pradesh. The sample was shade dried and pulverized to powder using a mechanical grinder. 500g of plant powder of the sample was soaked in ethanol for 72 h with intermittent shaking then filtered through Whit man No. 41 filter paper and concentrated under reduced pressure at 40°C using rotary evaporator to obtain a viscous semi solid mass/extract.

GC-MS Analysis

Gas-Chromatography Mass Specrometry (GC-MS) analysis of the ethanol extracts of *Mussaendamacrophylla*was carried out in Shimadzu GCMS-QP-2010 plus system. RTx-5 Sil MS column (30 m × 0.25 mm id × 0.25 film thickness) was used for the analysis. The operating conditions of the column were as follows:

Oven temperature program from 80°C to 210°C at 4°C/min withhold time of 2 min and from 210°C to 300°C at 15°C/min withhold time of 5 min, and the final temperature was kept for 20 min. The injector temperature was maintained at 270°C, the volume of injected sample was 0.3µl; pressure 85.4kPa, total flow 76.8mL/min, column flow 1.21 mL/min, linear velocity 40.5 cm/sec, purge flow 3.0 mL/min, split ratio: 60.0; ion source temperature 230°C; scan mass range of m/z 40-600 and interface line temperature 280°C. The identification of compounds was performed by comparing their mass spectra with data from NIST 11 (National Institute of Standards and Technology, US) and WILEY 8.

Identification of phytocompositions

The identification of compounds was performed by comparing their mass spectra with data from NIST 11 (National Institute of Standards and Technology, US) and WILEY 8.

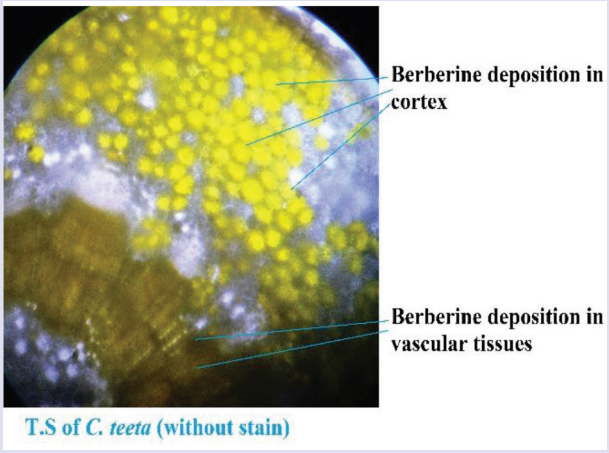


Figure 3: T.S of rhizome shows alkaloid deposition in Parenchymatous tissues.

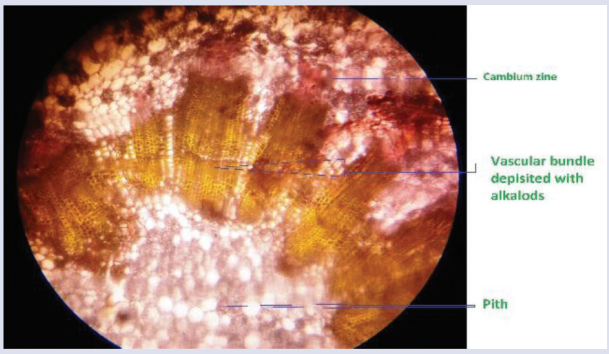


Figure 4: T.S of rhizome shows alkaloids deposition in vascular bundles.

RESULTS

Ethnobotany, distribution and threatening status

Mishmis and other tribes of Arunachal Pradesh use *C.teeta* in case of health problems like malaria, stomach ache, dysentery, Cold and Cough, Diarrhea, blood Dysentery, Typhoid, High blood pressure , Jaundice and to bring down body temperature during fever and also to relieve pain of eyes conjunctives. The rhizome is uprooted manually, hairs of rhizome are cleared, washed, sundried Figure 2 and grinded, soak in water then filtered and stored. In the word of Kanjilal and Borthakur.[15] Rhizome were brought in tiny little wicker baskets to Sadiya (Assam) where dealers from other Provinces go to buy them. Mishmis bartered M. teeta for opium with Meyor, an ethnic Tibetan tribe, now Arunachali after 1988.[19] They also brought the Yunnan Chinese silver.[20] Idu mishmi (Chulikatiya maanu) exchanged daos, musks, ornaments and salt for *Coptis teeta* with Assamese.[21] The open selling is illegal, but one can purchase at about Rs.2000/kg if source or seller can be located.

Dose and Preparation

The rhizome is up rooted; the uprooted rhizome is washed thoroughly in running water and dried under the sunlight (in case of immediate need, dried over the fire place). Dried rhizomes are either cut into pieces or grinded into powder form. These pieces or powder is soak in water (cold/lukewarm) for at least 30 min or until the solution appears yellow. For faster result the latter is recommended. The best way to avail its benefit is to consume the decoction twice a day before food.

Pharmacognostic studies

The rhizome is rough, live or fresh rhizome appears golden yellowish in colour while it looks brownish in dry; leaf scar is prominent and gives rhizome a rough and almost un-straight shape. Root stock profusely originate from the points where leaves developed and less profuse on underground rhizome part. The outermost surface of the transverse sectioned rhizome is covered with multi-layered cork upon cell wall; tissues are well differentiated into cortex, vascular bundles and pith. Alkaloids are chiefly deposited at cortex and Vascular bundles. Parenchymatous and conducting tissues are deposited with full of yellowish alkaloids while hardly any alkaloid deposition on pith Figure 3 and 4.

The powder studies of the rhizome show dull yellow powder, bitter on taste with total ash of at least 3.7 %, acid insoluble 0.8%, ethanol soluble extract of at least 15 % and water-soluble extract weigh 11% Table 1.

When powder was treated with different chemicals and observed under normal light and ultraviolet light following results were recorded. Powder as such was yellow in day light while it was bright yellow under UV light; dark brown in day light when treated powder with Sodium hydroxide but it appears dark green under UV light; powder was light red in day light when treated with acetic acid while it appears light green under UV light; powder was light brown in day light when treated with nitric acid in day light but it was light green under UV light; dark brown colour appeared in day light when treated with sulfuric acid but it was deep green when observed under UV light. Powder was turmeric colour in day light when treated with hydrochloric acid but dark green in UV light and the powder was brown in day light when treated with ferric chloride while it was dark green under UV light Table 2.

DISCUSSION

Coptis teeta is a an endemic and highly threatened medicinal plant, this herb is uprooted to collect the rhizome for medicinal purposes and put high pressure on their population, various methods to propagate shows partial success, protection and *in situ* conservation to increase population is important and further research on the line of pharmaceutical domain should be carried out to bring this useful herb to fight various health problems.

PHYTOCONSTITUENTS

A total of 56 compounds have been identified from the studied sample Table 3 and 5. Berbine, 13,13a-didehydro-9,10-dimethoxy-2,3-(methylenedioxy)- was found to cover highest areas in TIC peak report with the percentage of 41.08 % as given in Table 4. Other compounds like Deoxyaniflorine (17.04%), Indeno[1,2-b] quinoxalin-11-one, 2-methyl- (5.53%), Hexadecanoic Acid, Methyl Ester. (5.45 %), 1,1'-Biphenyl, 3,3',4,4',5,5'-hexamethoxy- (5.42 %), 9,12-Octadecadienoic acid (Z,Z)-, Methyl Ester (4.92 %), Stigmast-5-EN-3-OL, (3.BETA.)-, (1.90 %),

Table 1: *C. teeta* ash contents.

Parameter	Not less than (%)
Total ash	3.7
Acid insoluble	0.8
Ethanol soluble extractive	15
Water soluble extractive	11

Table 2: Florescence studies of *C.teeta* powder

Florescence study

C. teeta (rhizome powder)	Colour			
	Day light		Ultraviolet light	
Powder as such	Yellow		Light yellow	
Powder + NaOH	Dark brown		Dark green	
Powder + Acetic acid	Light reddish		Light green	
Powder + HNO_3	Light brown		Light green	
Powder + H_2SO_4	Dark brown		Dark green	
Powder + HCl	Turmeric colour		Dark green	
Powder + $FeCl_3$	Brown		Dark green	

Table 3: Compound table of *C.teeta* rhizome (ethanolic extract)

Peak	Retention time	Area	Area %	Name
1	4.543	665626	0.89	1,2,3, Propanetriol
2	5.499	72442	0.10	1-ethyl-2-pyroolidinone
3	8.035	61692	0.08	8-nonenoic-8,9-D2 acid, methyl ester
4	8.563	55865	0.07	DL-Valine, N-acetyl-, methyl ester.
5	8.890	269346	0.36	Benzenepropanoic acid, methyl ester.
6	9.208	104521	0.14	4,6-heptadeconoic acid,3,3,6-trimethyl -, methyl ester
7	9.341	462969	0.62	Bicyclo(5.2.0)Nonane, 4,8,8-trimethyl -2-methylene
8	9.518	1324264	1.76	2-methoxy-4-venylphenol
9	10.010	426988	0.57	Phenol,2,6-dimethoxy
10	10.227	193470	0.26	Cysteamine Sulfonic Acid
11	10.452	138093	0.18	2-propenoic acid, 3-phenyl, methyl ester, (Z)
12	11.888	82636	0.11	7-isopropophenyl-4A-Methyl-1-methylenedecahydronphthalene
13	11.971	104670	0.14	1,3-benzenedicarboxylic acid, dimethyl ester
14	12.112	57838	0.08	Nonanoic acid Methyl ester
15	12.233	66635	0.11	Benzoic acid, 4 Hydroxy-3-methoxy-, methyl ester
16	12.288	91994	0.12	Benzene, 1,2,3- Trrimethoxy-5-Methyl
17	12.795	314501	0.42	4-Methyl-2,5-Dimethoxy benzaldehyde
18	13.011	65210	0.09	Octane,3-5- dimethyl
19	14.471	131387	0.17	Heptadecanoic acid Methyl Ester
20	14.913	43682	0.06	4-(1E)-3-hydroxyl-1-Propenyl)-2-methoxyphenol
21	15.172	113044	0.15	Cyclopentanetridecanoic acid, Methyl ester.
22	15.268	84519	0.11	2-methyltetracosane.
23	15.400	57519	0.08	Dichloroacetic Acid, undec-2-enyl ester.
24	15.562	57846	0.08	Octadecanoic Acid, Methyl Ester.
25	16.364	54961	0.07	6-Octadecenoic Acid, Methyl Ester, (Z)-
26	16.409	320694	0.43	9-Hexadecenoic Acid, Methyl Ester, (Z)-
27	16.501	67474	0.09	9-Octadecenoic Acid, (Z)- Methyl Ester.
28	16.618	4092985	5.45	Hexadecanoic Acid, Methyl Ester.
29	17.055	1111411	1.48	Pentadecanoic Acid.

Continued...

Table 3: Cont'd.

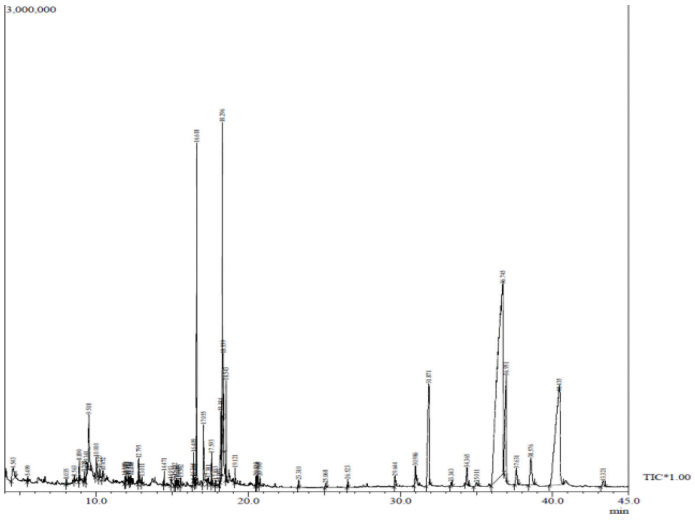
30	17.381	139026	0.18	Cyclopropanenonanoic acid, 2-[(2-butylcyclopropyl) Methyl]
31	17.593	324315	0.43	Hexadecanoic acid, 15-Methyl-, Methyl Ester.
32	17.833	58432	0.08	Hexadecane-1, 16-diol.
33	18.093	39891	0.05	Noroxyhydrastinine
34	18.184	907073	1.21	n-Nonadecanol-1
35	18.296	3699241	4.92	9,12-Octadecadienoic acid (Z,Z)-, Methyl Ester.
36	18.359	1105031	1.47	9,12,15-Octadecatrienoic acid, Methyl Ester(Z,Z,Z)-
37	18.543	982364	1.31	Methyl Streate
38	19.121	244706	0.33	(9E,12E)-9,12 Octadecadienoyl Chloride
39	20.513	130030	0.17	Eicosanoic Acid, Methyl Ester.
40	20.600	170007	0.23	Trans-2-Pinanol
41	20.750	149081	0.20	7-hydroxy, 6-methyl-bicyclo[4.3.0] nona-3-one
42	23.310	125676	0.17	Tetracosanoic acid, Methyl Ester
43	25.068	73666	0.10	Methyl Lignocerate
44	26.523	118431	0.16	Lignoceric Acid Methyl Ester
45	29.644	224927	0.30	1,2-Dimethoxy-4-[(1E)-1-Prophenyl]Benzene
46	30.986	470208	0.63	Canadine
47	31.871	4074513	5.42	1,1'-Biphenyl, 3,3',4,4',5,5'-hexamethoxy-
48	33.343	104062	0.14	alpha.-Tocopherol-.beta.-D-mannoside
49	34.365	543208	0.72	3,4-Quinolinedicarboxylic acid, 6-Methoxy-2-Phenyl-, Dimethyl ester
50	35.011	141443	0.19	N-(2,5-Di-Tertbutylphenyl)Phthalimide
51	36.745	31403799	41.78	Berbine, 13,13a-didehydro-9,10-dimethoxy-2,3-(methylenedioxy)-
52	36.745	4159109	5.53	Indeno[1,2-b]quinoxalin-11-one, 2-methyl-
53	37.631	616698	0.82	C-Chloro-N-[2-(6,7-Dimethoxy-Isoquinolin-1-YLMethyl)-4,5-Dimethoxy-Phenyl]-Methanesulfonamide
54	38.576	1427579	1.90	Stigmast-5-EN-3-OL, (3.BETA.)-
55	40.435	12810590	17.04	Deoxyaniflorine
56	43.321	220680	0.29	Cholest-4-en-3-one
		75158068	100.00	

Table 4: Compound activity

Peak No.	Area %	Name	Compound type	Activity
8	1.76	2-methoxy-4-venylphenol	Phenolic	Anti tumour[22], Antimicrobial Anti-inflammatory[23].
28	5.45	Hexadecanoic Acid, Methyl Ester.	Palmitic acid methyl ester	Antioxidant, Hypocholesterolemic, Nematicide, Pesticide, Antiandrogenic flavor, Hemolytic,5-Alpha reductase inhibitor*.
29	1.48	Pentadecanoic Acid.	Palmitic acid methyl ester	Antioxidant*.
34	1.21	n-Nonadecanol-1	Alcoholic compound	Anti-microbial[24].
35	4.92	9,12-Octadecadienoic acid (Z,Z)-, Methyl Ester.	Translinoleic acid	Hepatoprotective, antihistaminic, hypocholesterolemic, antieczemic*
36	1.47	9,12,15-Octadecatrienoic acid, Methyl Ester(Z,Z,Z)-	Linolenic acid ester	Antiinflammatory, Hypocholesterolemic, Cancer preventive, Hepatoprotective, Nematicide Insectifuge, Antihistaminic, Antieczemic, Antiacne, 5-Alpha reductase inhibitor Antiandrogenic, Antiarthritic, Anticoronary, Insectifuge*
37	1.31	Methyl Streate	Fatty acid methyl esters	Anti-diarrheal[25].
51	41.78	Berberine	Alkaloids	Antitumour[26], analgesic, Anesthetic, aantibacterial*.
52	5.53	Indeno[1,2-b]quinoxalin-11-one, 2-methyl-	spiroindenoquinoxaline Alkaloid	Anti cell proliferation of tumour cell[27].
54	1.90	Stigmast-5-EN-3-OL, (3.BETA.)-	Phytosterols	Potent anti-diabetic agent in regulating glucose transport[28].

https://phytochem.nal.usda.gov/phytochem/chemicals/show/14192?et=

Table 5: Chromatogram of C.teeta rhizome (ethanolic extract).



2-methoxy-4-venylphenol (1.76%), Pentadecanoic Acid (1.48%), 9,12,15-Octadecatrienoic acid, Methyl Ester (Z,Z,Z)- (1.47%), Methyl Streate (1.31 %), n-Nonadecanol-1 (1.21 %), Phenol,2,6-dimethoxy (0.57 %) respectively.

Phenolic, Palmitic acid methyl ester, Trans linoleic acid, Linolenic acid ester, Fatty acid methyl esters, Protoberberine alkaloids, spiroindeno quinoxaline, Phytosterols and alkaloids were the major compounds that has been reported to be biologically active from the studied sample Table 4. *Coptis teeta* contain compounds reported for activities like Anti tumour, Antimicrobial, Anti-inflammatory , Antioxidant, Hypocholesterolaemia, Nematicide, Pesticide, Antiandrogenic, flavor, Hemolytic, 5-Alpha reductase inhibitor, Hepatoprotective, antihistaminic, hypocholesterolaemia, antieczemic, Hypocholesterolaemia, Cancer preventive, Antihistaminic, Antieczemic, Antiacne, 5-Alpha reductase inhibitor, Antiandrogenic, Antiarthritic, Anti-coronary, Anti-diarrheal, Anti cell proliferation of tumour cell, Potent anti-diabetic agent in regulating glucose transport as given in Table 4.

CONCLUSION

It is concluded that *Coptis teeta* is distributed in the eastern Himalayas and a highly threatened important medicinal plant. The study gives hints that *Coptis teeta* contain at least 56 compounds, out of which ten compounds are biologically active against numbers of health problems like anti-cancer and anti-diarrhea. It is important to make out that the traditional use among the tribal people of Arunachal Pradesh for various health problems like anti-diarrhea, stomach pain and to bring down the temperature of fever could be correlated with the reported compounds present and found in this study; 2-methoxy-4-venylphenol as Anti-tumour.[22] Antimicrobial Anti-inflammatory[23] likewise Methyl Streate has already been reported as antidiarrheal.[25] Therefore, it is concluded that *Coptis teeta* need protection, propagation and further studies for the welfare of ecosystem and mankind.

ACKNOWLEDGEMENT

The author acknowledge O. Miso, G. Ering, M. Langkam, A. Tayang of BSc. final year Botany major students of J.N. College, Pasighat for their laboratory assistance, Mrs Y.J Lego, a local healer for materials and Dr. A Kumar, AIRF, JNU, New Delhi for GC-MS work.

CONFLICT OF INTEREST

The authors declare no conflict of interest.

ABBREVIATION USED

C.teeta: Coptis teeta Wall; **GCMS:** Gas Chromatograpy Mass Spectrometry.

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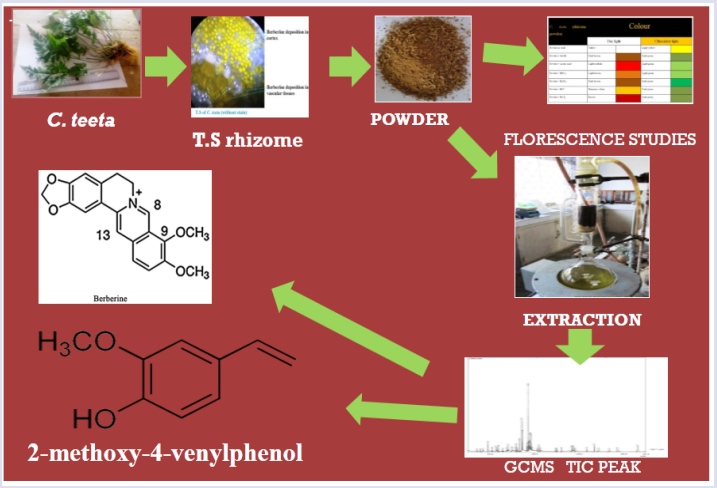
GRAPHICAL ABSTRACT



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Cite this article: Payum T. Distribution, Ethnobotany, Pharmacognosy and Phytoconstitents of *Coptis Teeta* Wall.: A Highly Valued and Threatened Medicinal Plant of Eastern Himalayas. Pharmacog J. 2017;9(6)Suppl:s28-s34.