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# PHYTOCONSTITUENTS OF *SPILANTHES OLERACEA* LINN. SHOOT: A MEDICINAL FOOD HERB USED AMONG TRIBAL PEOPLE OF ARUNCHAL PRADESH, INDIA

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**Keywords:**

Tribal People, Phytoconstituents, Traditional Knowledge, Toothache, Appetizer

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**ABSTRACT:** Herb is an indispensable food ingredient for the tribal people of Arunachal Pradesh. They practices innumerable traditional food recipe to maintain good health. *Spilenthes oleracea* L. is one of such medicinal food plant used by tribal people of Arunachal Pradesh. The study was carried out to find out the phytoconstituents of *S. oleracea* shoot. Ethanolic extract of the sample was subjected to GC-MS to know the Phytoconstituents of this herb and correlate the phytoconstituents and health claim thereof if any. This study reports the presence of 48 phytocompounds and a total of 10 compounds identified from this sample have been reported to show painkiller effect that complements the traditional knowledge on this herb among the tribal people of the study site and scientific evidence. Sesquiterpene, Polyenoic fatty acid, Steroidal, Palmitic acid ethyl ester and Hydrocarbon are the major group of compound found from the sample. Present work advances *S. oleracea* as a medicinal food herb.

**INTRODUCTION:** The environment around has been influencing man's culture since the dawn of time and he started to make use of available resources around him from antiquity. Man must have learnt the use of plant on 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]. Therefore, Knowledge on the therapeutic properties of plant is predated recorded history [2]. There are several historical indications that claim the ancient use of natural remedies to resolve primary health problems in different part of the world in different forms like rhymes, folk, Songs, symbols, inscriptions, paintings, Cueniform tablet, Ayurvedic texts, Papyrus scroll, Herbals, etc. [1, 2]. Food without Dal, Wheat, Potato, Oil and Spice is the characteristic features of the indigenous food system in Arunachal Pradesh [3]. Let food be your medicine, once said Hippocrates (c. 460 – c. 370 BC) over 2500 years' ago [4]. Such medicinal food concepts and practice is still observable in the indigenous food systems among indigenous people in various pockets of the world in general and Arunachal Pradesh in particular [3]. Further, in the word of Etkin and Ross [5], wild plants that are retained in local food cultures are inseparable from traditional therapeutic systems and Pieroni and Price [6] remarked that it is difficult to draw a line between food and medicine; food may be medicine and medicine may be food. Marriot Bernadette [7] also marked that links of diet and health are no longer questioned. Arunachal Pradesh, the "land of the dawn-lit mountains", which is also known as the "Paradise of the

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Botanists in India is also known for her rich biodiversity [7, 8]. This remote state was first Botanized by Griffith in 1836 only. The state harbors thousands of plants with equal numbers of traditional uses. The indigenous people use many plants as food as well as medicine also but these traditional practices are yet to be validated scientifically. The traditionally used medicinal food plants may contain various useful compounds if studied scientifically and may become boon for the mankind on scientific intervention. Hence *Spilenthes oleracea* has been taken up in the present study as this herb is used as vegetables, appetizer as well as to relief toothache in the studied area.

Numerable workers have carried out research on different aspects of *Spilanthes* species. The genus of *Spilanthes* is composed of about 60 species, distributed in the tropics and the subtropics. These plants are an annual herb that grows up to 60 cm. The plant has yellowish red head flowers. *Spilanthes* species as an Ornamed plant with ornamental and medicinal value in peninsular India [9]. Traditionally this herb is used in fish poisoning, gastric problem and in blood clothing among the Monpas of Arunachal Pradesh [10]. Alkylamides has also been isolated from another related *Spilathes* species collected from Taiwan [11]. Pharmacological works on *Spilathes acmella* is also well reviewed [12]. Recently spilanthol from *Acmella olerceae* has been reported to be effective as diuretic in Urine concentration mechanism to lower the intracellular level of cAMP impairing NKCC2 phosphorylation and waerchannel AQP2 membrane expression in mouse kidney [13]. *Spilanthes acmella* has also been reported to useful in the treatment of rheumatoid arthritis, gastro intestinal disturbances, treatment of malarial and fungal infections [14, 15]. It is more or less evident that *Spilanthes* species are useful in the treatment of toothache but not much work on the phytoconstituents of the selected species has been done particularly from Arunachal Pradesh.

With above backdrops, the shoot of *Spilathes oleracea* has been studied to know the phyto-constituents of this plant by using GC-MS. Because, 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 [16]. Further, Gas chromatography-mass spectrometry (GC-MS) is the most commonly used technique for the analysis of lipo-soluble constituents, especially volatile / semi-volatile compounds, and their metabolites in biological fluids due to its high resolution, selectivity and sensitivity [17].

## MATERIAL AND METHODS:

**Plant Material:** *Spilanthes oleracea* Linn. a trailing herb, opposite and triangular leaves, ovate or lanceolate, distantly serrate, acute, 3 nerved. Head yellow, solitary or in long terminal panicles; several series of outer flowers with small rays, inner all tubular; involucres ovoid or campanulate; breacts sub-2 seriate, slightly unequal. Receptacle elongate; pales enclosing the flowers. Antherbase truncate. Style-arms rather long, truncate. Fruit of disc flowers usually compressed. Pappus absent or of 2-3 bristles [18]. The material was bought from Pasighat local market, East Siang District of Arunachal Pradesh, India in the September month of 2016.

**Preparation of Extract:** The sample was shade dried and pulverized to powder using a mechanical grinder. 600g of plant powder was soaked in ethanol for 72 hours with intermittent shaking then filtered through Whatmann No. 41 filter paper and concentrated by using water bath.

**GC-MS Analysis:** Gas-Chromatography Mass Specrometry (GC-MS) analysis of the ethanol extracts of *Croton tiglium* carried out in Shimadzu GCMS-QP-2010 plus system. RTx-5 Sil MS column (30 m X 0.25 mm id X 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 40.5cm/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 Phytocompounds:** 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.

**RESULTS AND DISCUSSION:** The ethanolic extract GCMS chromatogram of *S. oleracea* L. (**Fig. 1**) shows 48 peaks indicating the presence of at least forty eight phytochemical constituents about 15 percent of the phytoconstituents are essential oils. On comparison of the mass spectra of the constituents with the NIST 11 library and Willey 8 library, the forty eight phytocompounds were characterized and identified as given in **Table 1**. Compounds related to the traditional uses *i.e.* anaesthetic and corresponding reported biological activities are given in **Fig. 2, 3, 4** and **5**. Selected twenty compounds that have reported to be biologically active are given in **Table 2**.

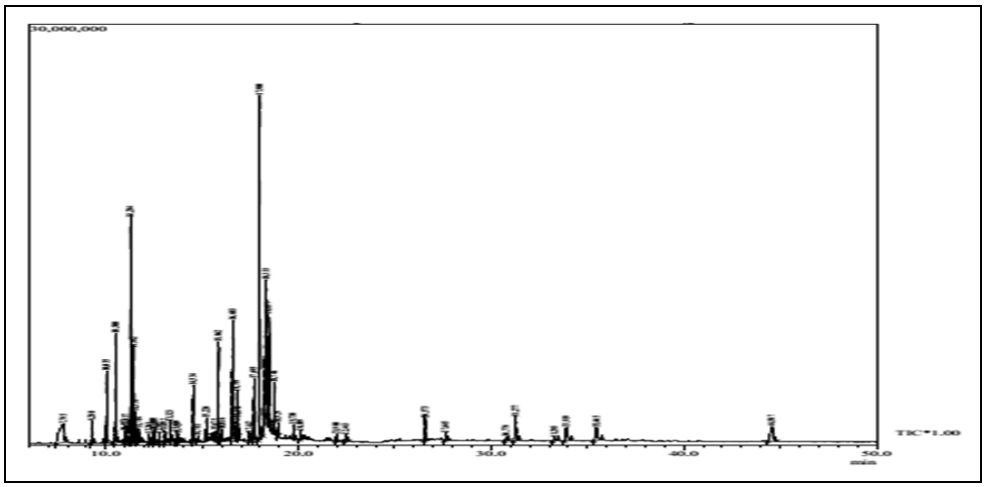

**FIG. 1: GCMS ETHANOLIC EXTRACT SPECTRA OF *SPILANTHES OLERACEA***

**TABLE 1: TIC PEAK REPORT**

| Peak | R. time | Area | Area % | Compound |
|---|---|---|---|---|
| 1. | 7.795 | 23911441 | 5.65 | 1-Butanol, 3-methyl-, acetate |
| 2. | 9.280 | 3460491 | 0.82 | .delta.- elemene |
| 3 | 10.035 | 8021597 | 1.90 | Beta-elemen-(2) |
| 4 | 10.500 | 13863432 | 3.28 | Trans (.Beta.)-Caryophyllene |
| 5 | 10.957 | 1232917 | 0.29 | .alpha.- humulene |
| 6 | 11.037 | 1852716 | 0.44 | Cyclododecene, (Z)- |
| 7 | 11.284 | 45087357 | 10.65 | Cedrene |
| 8 | 11.442 | 13920026 | 3.29 | 1,3,6,10-Dodecatetraene,3,7,11-Trimethyl- |
| 9 | 11.531 | 3270641 | 0.77 | Beta-Bisabolen |
| 10 | 11.704 | 1308043 | 0.31 | Gamma Muurolen |
| 11 | 12.261 | 1185027 | 0.28 | 1,2,3,5,6,7,8,8a-Octahydro-8-Methyl-2-(1-methylethyl)-(4H) 1,4a-(epoxymethano)napthalen-4-one |
| 12 | 12.488 | 2187947 | 0.52 | E-14-Hexadecenal |
| 13 | 12.557 | 1533126 | 0.36 | Caryophyllene, Epoxide |
| 14 | 12.782 | 1142179 | 0.27 | .beta.-oplopenone |
| 15 | 13.052 | 1445335 | 0.34 | (-)-Spathulenol |
| 16 | 13.323 | 2104444 | 0.50 | 10,12-Hexadecadien-1-ol |
| 17 | 13.656 | 1154473 | 0.27 | E-15 heptadecenal |
| 18 | 13.729 | 1884719 | 0.45 | (-)-.beta.-Caryophyllene Epoxide |
| 19 | 14.534 | 9025751 | 2.13 | 1,3-Bis(Cinnamoyloxymethyl) adamantane |
| 20 | 14.761 | 547229 | 0.13 | 1-Octadecene |
| 21 | 15.226 | 1888490 | 0.45 | Neophytadiene |
| 22 | 15.675 | 1200858 | 0.28 | 3,7,11,15-Tetramethyl-2-hexadecen-1-Ol |
| 23 | 15.862 | 19640443 | 4.64 | N-Isobutyl-10-(Isobutylcarbonyloxy)(2E,6Z,8E)- |

| | | | | |
|---|---|---|---|---|
| | | | | Decetrienamide |
| 24 | 16.016 | 1668789 | 0.39 | .gamma.-dodecadienolactone(Z-Z)) |
| 25 | 16.603 | 37417062 | 8.84 | Pentadecanoic acid |
| 26 | 16.745 | 1380295 | 0.33 | 1,3-Methanopantalene, 1,2,3,5-tetrahydro- |
| 27 | 16.799 | 4917398 | 1.16 | Hexadecanoic acid, ethyl ester |
| 28 | 16.870 | 1950113 | 0.46 | 2-Sec-Butyl-4,6-Dinitrophenyl 3-Methyl-2-Butenoate |
| 29 | 17.442 | 1103950 | 0.26 | 2-Propenal, 2-Methyl-3-Phenyl- |
| 30 | 17.695 | 12143006 | 2.87 | N-Isobutyllundeca-2 (E)-en-8,10-dynamide |
| 31 | 17.980 | 68250177 | 16.13 | 3,7,11,15-Tetramethylhaxadec-2-EN-1-OL |
| 32 | 18.333 | 60112311 | 14.20 | 9,12,15-Octadecatrienoic acid, (Z,Z,Z)- |
| 33 | 18.393 | 5696115 | 1.35 | Ethyl (9Z, 12Z)-9,12-Octadecadienoate |
| 34 | 18.457 | 12055006 | 2.85 | Ethyl Linoleate |
| 35 | 18.746 | 5446697 | 1.29 | N-Isobutyl-(2E, 4Z,8Z,10E)-dodecatetraenamide |
| 36 | 18929 | 1321338 | 0.31 | Bicyclo[2.1.0]pentane,1,4-dimethyl |
| 37 | 19.730 | 1346230 | 0.32 | 1,3-Bis(cinnamoyloxymethyl)adamantane |
| 38 | 20.109 | 2031572 | 0.48 | Fenaclon |
| 39 | 21.980 | 1280014 | 0.30 | Eicosane |
| 40 | 22.443 | 1980649 | 0.47 | Benzene, (2,2,-dimethylcyclopentyl)- |
| 41 | 26.573 | 3930417 | 0.93 | Squalene |
| 42 | 27645 | 1589180 | 0.38 | Tetratetracontane |
| 43 | 30.799 | 1189878 | 0.28 | Tetracontane |
| 44 | 31.277 | 7787426 | 1.84 | dl-.alpha.-Tocopherol |
| 45 | 33.295 | 1800130 | 0.43 | Ergost-5-en-3-ol,(.Beta.)- |
| 46 | 33.919 | 7056383 | 1.67 | Stigmasta-5, 22-dien-3-Ol |
| 47 | 33.465 | 7574891 | 1.79 | Stigmast-5-en-3-Ol, (3- beta.)- |
| 48 | 44.587 | 11287705 | 2.67 | Phytol, acetate |
| | | 423185414 | 100.00 | |

The most prevailing compounds were 3, 7, 11, 15-Tetramethylhaxadec-2-En-1-Ol (16%), 9, 12, 15-Octadecatrienoic acid, (Z, Z, Z)- (14%), Cedrene (10.65%), Pentadecanoic acid (8%), 1-Butanol, 3-methyl-, acetate (5.65%), N-Isobutyl-10-(Isobutyl-carbonyloxy) (2E, 6Z, 8E) -Decetrienamide (4.64%), Trans (Beta)- Caryophyllene (3.28%), N-Isobutyllundeca -2 (E)-en-8, 10-dynamide (2.87%), Ethyl Linoleate (2.85%), Phytol, acetate (2.67%), 1, 3- Bis (Cinnamoyloxy-methyl) adamantane (2.13%) Beta- elemen- (2) (1.90%) dl- alpha-Tocopherol (1.84%), Stigmast-5-en-3-Ol, (3- beta)- (1.79%), Ethyl (9Z, 12Z)- 9, 12-Octadecadienoate (1.35%) and N-Isobutyl-(2E, 4Z, 8Z, 10E)- dodeca-tetraenamide (1.29%). Sesquiterpene, Polye-noic fatty acid, Steroidal, Terpene alcohol, Palmitic acid ethyl ester and Hydrocarbon were the major group of compound found from the sample.

**TABLE 2: BIOLOGICAL ACTIVITY OF COMPOUNDS**

| Sl. No. | Compound | Activity |
|---|---|---|
| 1 | (-)-beta-Caryophyllene Epoxide | Anti-cancer, anti-inflammatory, antibiotic, antioxidant and local anaesthesis activities [19] |
| 2 | (-)-Spathulenol | Sedative and analgesic [20] |
| 3 | alpha- humulene | Anti-inflammatory [21] |
| 4 | delta- elemene | Anti-proliferative [22] |
| 5 | .gamma-dodecadienolactone (Z-Z)) | Antimicrobial [23] |
| 6 | 1-Octadecene | Lubricants*** |
| 7 | 3,7,11,15-Tetramethylhaxadec-2-EN-1-OL | Anti-inflammatory and antimicrobial** |
| 8 | 9,12,15-Octadecatrienoic acid, (Z,Z,Z)- | Antiinflammatory, Hypocholesterolemic, Cancer preventive, Hepatoprotective, Nematicide, Insectifuge Antihistaminic, Antiarthritic, Anticoronary, Antieczemic, Antiacne, 5-Alpha reductase inhibitor Antiandrogenic ** |
| 9 | dl-alpha-Tocopherol | Anti-inflammatory, antioxidant, antimicrobial, radical scavenging, antispasmodic** |
| 10 | Ergost-5-en-3-ol, (Beta)- | Anti-microbial, Anti-inflammatory effect** |
| 11 | Ethyl Linoleate | Antidiabetic [24] |
| 12 | Hexadecanoic acid, ethyl ester | Antioxidant, Hypocholesterolemic, Nematicide, Pesticide, Lubricant, Antiandrogenic, Flavor [25] |

| | | |
|---|---|---|
| 13 | Neophytadiene | Antipyretic, analgesic and anti-inflammatory, antimicrobial, antioxidant** |
| 14 | Pentadecanoic acid | Antioxidant** |
| 15 | Squalene | Antioxidant, antitumor, immunostimulant, diuretic [24] |
| 16 | Stigmast-5-en-3-Ol, (3- beta.)- | Analgesic, antidiabetic, Anti-inflammatory** |
| 17 | Stigmasta-5, 22-dien-3-Ol | Anti-bacterial, Anti-inflammatory, Antiarthritic,antiasthma, diuretic** |
| 18 | Tetracontane | Antibiotic [26] |
| 19 | Tetratetracontane | Antioxidant [27] |
| 20 | Trans (Beta)-Caryophyllene | Antileishmanial [28] |

**Dr. Duke's Phytochemical and Ethnobotanical Databases.
*** (https://pubchem.ncbi.nlm.nih.gov/compound/1octadecene#section=Consumer-Uses

(-)-beta-Caryophyllene Epoxide is anticancer, anti-inflammatory, antibiotic, antioxidant and local anaesthetic activities [19], (-)-Spathulenol is an anesthetic and sedative agent [20], alpha- humulene is an anti-inflamatory [21], delta- elemene is an anti-proliferative compound [22], gamma-dodecadieno-lactone (Z-Z) is antimicrobial [23], 1-Octadecene is a lubricant*** 3, 7, 11, 15-Tetramethylhaxadec-2-EN-1-OL is an anti-inflammatory and anti-microbial**, 9, 12, 15-Octadecatrienoic acid, (Z,Z,Z)- is anti-inflammatory, hypocholesterolemic, cancer preventive, hepatoprotective, nematicide, insectifuge antihistaminic, antiarthritic, anticoronary, antieczemic, antiacne, 5-Alpha reductase inhibitor antiandrogenic**, dl-alpha-Tocopherol is anti-inflammatory, antioxidant, antimicrobial, radical scavenging, antispasmodic**, Ergost-5-en-3-ol,(Beta)- is anti-inflammatory and antimicrobial**, Ethyl Linoleate is antidiabetic [24], Hexadecanoic acid, ethyl ester is antioxidant, hypo-cholesterolemic, nematicide, Pesticide, Lubricant, Anti-androgenic, Flavor [25], Neophytadiene is antipyretic, analgesic, and anti-inflammatory, antimicrobial, antioxidant**.

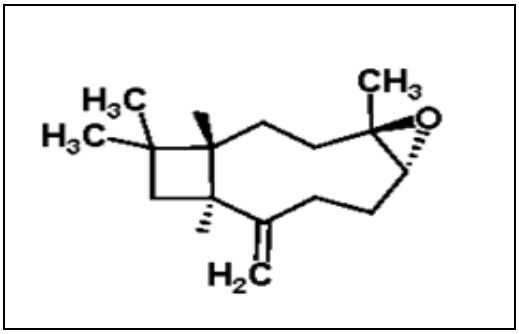


**FIG. 2: (-)-BETA-CARYOPHYLLENE EPOXIDE**

Pentadecanoic acid is an antioxidant, Squalene is antibacterial, antioxidant, antitumor, cancer preventive, immuno-stimulant, chemo preventive, lipoxygenase inhibitor, pesticide, diuretic [24], Stigmast-5-en-3-Ol,(3-beta)-is analgesic, antidiabetic and anti-inflammatory**, Stigmasta-5, 22-dien-3-Ol is anti-oxidant, antibacterial activity, antiinflamma-tory, antiarthritic antiasthma, diuretic**, Tetracon-tane is antibiotic [26], Tetratetracontane is an anti-oxidant [27], Trans (Beta)-Caryophyllene is an anti-leishmanial [28].

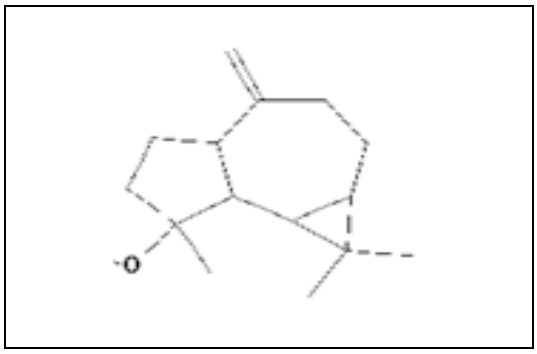


**FIG. 3: (-)-SPATHULENOL**

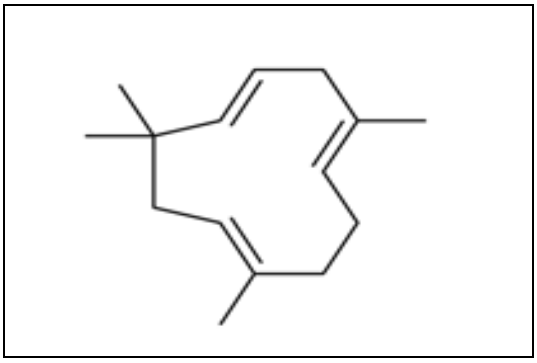

**FIG. 4: ALPHA- HUMULENE**

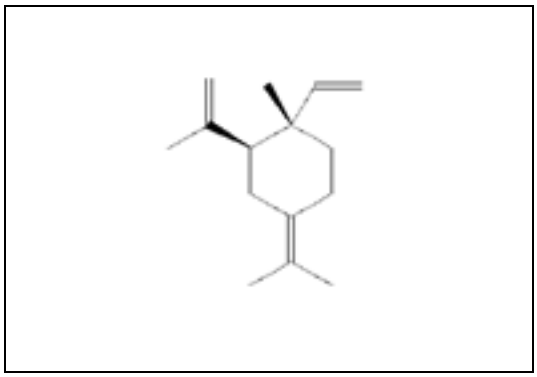

**FIG. 5: DELTA- ELEMENE**

**CONCLUSION:** From this work, it could be concluded that *S. oleracea* shoot contains various useful phytocompounds for numbers of health problems like inflammation, cancer, diabetic and antioxidation, antbacterial etc., Out of forty eight compounds, a total of ten compounds identified from this sample has been reported to show painkiller effect by various workers referred in table number (2) that complements the traditional knowledge on this herb among the tribal people of the study site and scientific evidence. In addition, few compounds that act as an antioxidants and anti cancers are also present in this herb. The study felt the need of further scientific studies that could lead in the discovery of potent useful medicines for various health problems and anaesthetic in particular.

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**CONFLICT OF INTEREST:** The author doesn't declare any conflict of interest.

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