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Proximate composition and GC-MS analysis of ethanol extract of *Solanum spirale* Roxb.

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ABSTRACT

Solanum spirale Roxb. is abundantly grown in eastern Himalayan regions. The shoot is used as a vegetable as well as medicine to control high blood pressure. The present study was carried out to profile the phytochemical compositions, mineral contents and proximate composition of the *Solanum spirale* shoot. GC-MS was used for the identification of phytochemicals and methods described in standard protocols were used to study minerals and proximate composition. A total of 40 phytochemicals were recorded from the ethanol extract. Mineral contents of the sample 0.115 (Fe), 0.07 (Mn), 0.015 (Cu), 0.040 (Zn), 2.25 (Mg), 3.08 (Na) and 16.7 (K) mg/g while proximate composition were 76.25 ± 0.093 % (moisture) 3.82 ± 0.26 % (Carbohydrate), 12.54 ± 0.08% (Total ash), 0.39 ± 0.98% (Crude protein), 6.12 ± 1.07 % (Crude fibre), 0.37 ± 0.07 % (Crude fat). (Z,Z)-6,9-Cis-3,4-epoxy-nonadecadiene occupied highest area percentage in TIC peak report with 24.55%, followed by Pentadecanoic acid with 18.81 percent, with Pentadecane with 9.99%, Hexadecanoic acid, Ethyl ester with 8.64%, (2E)(7R,11R)-3,7,11,15-Tetramethylhexadec-2-en-1-ol with 7.75%. Useful phytochemicals related to health problems including anticancer, anti-inflammatory, antioxidant, antitumor, cardioprotective, hypocholesterolemic, increase zinc bioavailability, inhibit uric acid formation, antibacterial, anti-inflammatory, antioxidant etc. are major components and such findings advocate *Solanum spirale* as a nutraceutical herb.

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INTRODUCTION

Food and medicine represent a continuum rather than artificial categories; Overlapping nature of traditional food system and medicine lead to the investigation of phytochemicals that explains the food culture and health outcomes [1]. Any of the edible wild plants that are included in local food baskets have both therapeutic and dietary functions and such medicinal foods have been part of Eastern Medicinal theories since ancient times; In Traditional Chinese Medicine also the concept of food as medicine is based on the same basic theories and they are vice-versa [2,3]. North Eastern States of Eastern Himalayas is rich in biodiversity [4,5], because of diverse ethnic groups, the region is equally rich in culture, traditional practices and customs. These people have rich knowledge on the utility of plant resources including herbal remedy; the region has many dimensions in food habits and flavour [6]. The shoot of *Solanum spirale* used as food as well as medicine by the tribal people in Eastern Himalayan region; as a medicine, it is used to lower down high

blood pressure, loose motion and stomach pain, tender shoot is steam cooked as vegetable and berry is used as appetizer. The present study was selected to discourse the phytochemicals and proximate composition of *Solanum spirale*. This herb also grows commonly in North East India, China and Thailand. It blooms with spirally arranged white flower and berries are orange on ripe. Bark, leaf and berry tastes bitter. *S. spirale* has been reported to be an antioxidant potential herb [7]. The herb is also used as anti-tussive and anti-inflammatory agent historically [8]. Primary metabolites like proximate composition and minerals in a diet play vital role in promoting health growth and development; Minerals regulate cell function in more than one way; they are constituents of skeletal structures, they maintain colloidal state of the body matter and regulate acid base equilibrium, diffusion, and osmotic pressure and they are components or an activator of enzymes. Proximate composition of a diet provides energy, protein and other requirements of a body for healthy growth and development; secondary metabolites help in the remedy of various health problems and work as a nutraceutical food. With

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the above backdrops, the present research was taken up to study the proximate composition, phytoconstituents and minerals present in *Solanum spirale* shoot.

MATERIAL AND METHODS

Sample Collection and Preparation

Solanum spirale shoots were collected from Renging Village of Arunachal Pradesh, India. The herbarium was deposited in the Department of Botany, JN.College herbarium section with voucher number JNCTP/SOL/092019. The sample was washed thoroughly in distilled water and dried in shade till the weight did not changed further; and pulverized into powder, using a mechanical grinder. 500g sample powder was subjected to Soxhlet apparatus in ethanol solvent, the slurry extract collected from rotary evaporator was used in the GC-MS analysis.

Proximate and Minerals Studies

For moisture study, fresh collected sample was used before drying. The following standard methods were followed in the proximate and minerals studies [9,10,11]. Ash content was determined in silica crucibles by incineration in a muffle furnace at 550°C for 5 hrs. Crude lipid was extracted by continuous Soxhlet extraction with petroleum ether (40-60°C). Crude fibre content was estimated by acid-base digestion with 1.25% H₂SO₄ and 1.25% NaOH solution. Nitrogen was estimated by KjeldelMethod with steam distillation and titrated with standard 0.01 M HCL solution. Crude protein content was estimated by multiplying the sample per cent Nitrogen content by a factor 6.25. (% protein = % Nitrogen X 6.26). Carbohydrate was estimated by Anthrone Method with an ultraviolet-visible (UV-Vis) spectrophotometer (Lamda-25, Perkin Elmer, Cambridge UK). Minerals were analysed from solution obtained when 1.0g of the samples were digested with concentrated 10ml nitric acid and kept overnight and heated till fumes of HNO₃ and allowed to cool and add 4ml concentrated Perchloric acid and heated again till clear solution is obtained and filtered into 100ml ml standard flask and made to mark with distilled water and analysed in atomic absorption spectrophotometer (Buck scientific model 200A).

Phytochemicals Studies

To study phytochemical compositions, All the solvents used were purchased from Merck Company analytical grade. GC-MS: Gas-Chromatography Mass Specrometry (GC-MS) analyses of the each extract were 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 40.5 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 NIST05 (National Institute of Standards and Technology, US) and WILEY 8.

RESULTS

A total of forty compounds (Figure 1) were identified (Table 1) from the ethanol extract and adequate mineral contents and proximate composition from *Solanum spirale* shoot was recorded; In the compound table, (Z,Z)-6,9-

Table 1: Chemical composition of ethanol extract of *Solanumspirale*

Peak	RT	Area %	Name of the compound
1	4.860	1.05	2-Pyrrolidinone, 1-methyl-
2	6.484	0.62	2,3-Dihydro-3,5-dihydroxy-6-methyl-4h-pyran-4-One
3	7.124	0.14	1-Tridecene
4	9.961	0.35	1-Tetradecene
5	10.091	0.38	alpha.-Tetradecene
6	11.876	0.15	2(4H)-Benzofuranone, 5,6,7,7a-tetrahydro-4,4,7a-trimethyl-
7	12.248	0.17	Fumaric acid, ethyl 2-isopropylphenyl ester
8	12.461	0.52	Phthalic acid
9	13.238	0.49	Piperidine, 1-(1-cyclopenten-1-yl)-
10	13.447	0.33	5-Hydroxymethyl-1,3,3-trimethyl-2-(3-methyl-buta-1,3-dienyl)-cyclopentanol
11	13.578	0.22	Santalol, cis, alpha.-
12	13.647	0.26	Spirio-10-(2,11-dioxabicyclo[4.4.1]undeca-3,5-diene)-2'-(oxirane), 1,3,7,7-tetramethyl-
13	14.307	0.18	6-(1-Hydroxyethyl)-2-iodo-4-oxa-tricyclo[4.2.1.0(3,7)]nonan-5-one
14	14.597	0.11	Benzyl Benzoate
15	14.637	0.22	Calendin
16	15.223	0.82	2,6,10-Trimethyl,14-ethylene-14-pentadecne
17	15.479	0.31	Cyclopropanenonanoic acid, 2-[(2-butylcyclopropyl)methyl]-, methyl ester
18	15.674	0.48	3,7,11,15-Tetramethylhexadec-2-en-1-ol
19	16.543	18.81	Pentadecanoic acid
20	16.714	0.16	Ethyl 9-hexadecenoate
21	16.797	8.64	Hexadecanoic acid, ethyl ester
22	17.746	0.17	Heptadecanoic acid, ethyl ester
23	17.938	7.75	(2E)(7R,11R)-3,7,11,15-Tetramethylhexadec-2-en-1-ol
24	18.240	24.55	(Z,Z)-6,9-CIS-3,4-Epoxy-nonadecadiene
25	18.381	3.75	Ethyl (9z,12z)-9,12-octadecadienoate
26	18.435	9.99	Dichloroacetic acid, tridec-2-ynyl ester
27	18.489	0.38	9-Octadecenoic Acid (Z)-
28	18.669	4.56	Octadecanoic acid, ethyl ester
29	18.961	0.44	1,E-11,Z-Octadecatriene
30	20.295	0.79	9-Octadecenoic acid (z)-
31	20.465	0.96	1,3-Cyclopentanedione, 4-hydroxy-2-pentyl-
32	20.611	0.79	Heptadecanoic acid, ethyl ester
33	23.457	0.51	Ethy L margarate
34	25.158	0.22	Octadecanoic acid, ethyl ester
35	26.357	0.22	Heptadecanoate
36	26.567	2.07	Squalene
37	29.844	3.39	.gamma.-Tocopherol
38	31.249	2.13	dl-.alpha.-Tocopherol
39	33.896	1.07	Stigmasta-5,22-dien-3-ol
40	35.436	1.89	Stigmast-5-en-3-ol, (3.beta.)-
		100	

Table 2: Biological activity of the identified compounds

Name of the compounds	Biological activity
2-Pyrrolidinone, 1-methyl-	Catechol-O-Methyl- transferase –inhibitor [12].
2,3-DIHYDRO-3,5-DIHYDROXY-6-METHYL-4H-PYRAN-4-ONE	Methyl-guinidine –inhibitor [12].
alpha.-Tetradecene	5-alpha-reductase inhibitor [12].
2(4H)-BENZOFURANONE, 5,6,7,7A-TETRAHYDRO-4,4,7A-TRIMETHYL-	Antidote, anti HIV integrase, HIF1 alpha inhibitor[12].
Fumaric acid, ethyl 2-isopropylphenyl ester	Smart drug, 11B-HSD inhibitor, 5 HETE inhibitor[12].
PHTHALIC ACID	Inhibit uric acid formation [12].
Santalol, cis-alpha.-	5-alpha-reductase inhibitor [12].
PENTADECANOIC ACID	Inhibit uric acid formation [12].
HEXADECANOIC ACID, ETHYL ESTER	Inhibit uric acid formation [12].
(2E)(7R,11R)-3,7,11,15-TETRAMETHYLHEXADEC-2-EN-1-OL	Endocrinoprotective [12].
(Z,Z)-6,9-CIS-3,4-EPOXY-NONADECADIENE	Increase zinc bioability [12].
ETHYL (9Z,12Z)-9,12-OCTADECADIENOATE	Increase zinc bioability [12].
Dichloroacetic acid, tridec-2-ynyl ester	Inhibit uric acid formation [12].
9-OCTADECENOIC ACID (Z)-	Acidifier [12].
OCTADECANOIC ACID, ETHYL ESTER	Inhibit uric acid formation [12].
9-OCTADECENOIC ACID (Z)-	Inhibit uric acid formation [12].
1,3-Cyclopentanedione, 4-hydroxy-2-pentyl-	17-beta-hydroxysteroid dehydrogenase-inhibitor [12].
Heptadecanoic acid, ethyl ester	Inhibit uric acid formation [12].
Ethy	Anticancer, antioxidant [12].
L margarate	
Squalene	Monooxygenase-inhibitor[12], antioxidant, antitumour[13].
gamma.-Tocopherol	Antioxidant, Tocopherol synergist, PPAR-Gamma-Antagonist [12].
dl.-alpha.-Tocopherol	Antioxidant, TNF-alpha inhibitor [12].
Stigmasta-5,22-dien-3-ol	Antimicrobial [14].
Stigmast-5-en-3-ol, (3.beta.)-	Anti-diabetic; apart from its existing cholesterol lowering efficacy [15].

CIS-3,4-epoxy-nonadecadiene was recorded to occupy the highest area percentage with 24.55% followed by Pentadecanoic acid with 18.81%; then Dichloroacetic acid, tridec-2-ynyl ester with 9.99%, while Hexadecanoic acid, ethyl ester was in 8.64%, Hexadecanoic acid, (2E) (7R,11R)-3,7,11,15-Tetramethylhexadec-2-en-1-ol with 7.75%, and other phytochemicals are gamma-Tocopherol, 2-Pyrrolidinone, 1-methyl-, 2,3-dihydro-3,5-dihydroxy-6-methyl-4h-pyran-4-one, 1-Tridecene, 1-Tetradecene, alpha.-Tetradecene, 2(4H)-benzofuranone, 5,6,7,7A-tetrahydro-4,4,7a-trimethyl-, Fumaric acid, ethyl 2-isopropylphenyl ester, Phthalic acid, piperidine, 1-(1-cyclopenten-1-yl)-, Santalol, cis-alpha.-, 5-Hydroxymethyl-1,3,3-trimethyl-2-(3-methyl-butyl-1,3-dienyl)-cyclopentanol, Spirio-10-(2,11-dioxabicyclo[4.4.1]undeca-3,5-diene)-2'-(oxirane), 1,3,7,7-tetramethyl-, Benzyl Benzoate, 6-(1-Hydroxyethyl)-2-iodo-4-oxa-tricyclo[4.2.1.0(3,7)]nonan-5-one, CALENDIN, 2,6,10-Trimethyl,14-Ethylene-14-Pentadecene, Cyclopropanenonanoic acid, 2-[(2-butylcyclopropyl) methyl]-, methyl ester, 3,7,11,15-tetramethylhexadec-2-en-1-ol, ethyl 9-hexadecenoate, Heptadecanoic acid, ethyl ester, (2e)(7r,11r)-3,7,11,15-tetramethylhexadec-2-en-1-ol, ethyl (9z,12z)-9,12-octadecadienoate, 9-octadecenoic acid (z)-, Octadecanoic acid, ethyl ester, 1e-11,z-octadecatriene, 9-octadecenoic acid (z)-, ethyl margarate, Heptadecanoic acid, ethyl ester, 1,3-cyclopentanedione, 4-hydroxy-2-pentyl-, octadecanoic acid, ethyl ester, Heptadecanoate. Mineral contents of the sample was recorded in mg/g as Fe 0.115, Mn 0.07, Cu 0.015, Zn 0.040, Mg 2.25, Na 3.08 and K 16.7 and proximate composition was recorded as 76.25 ± 0.093 % (moisture), 3.82 ± 0.26 % (Carbohydrate), 12.54 ± 0.08% (Total ash), 0.39 ± 0.98% (Crude protein), 6.12 ± 1.07 % (Crude fibre), 0.37 ± 0.07 % (Crude fat).

DISCUSSION

The *Solanum spirale* shoot contains useful primary as well as secondary metabolites. The ethanol extract of *Solanum spirale* shoot contains as many as twenty three useful phytochemicals (Table 2) in addition to proximate and mineral composition for healthy growth and development of body. (Z,Z)-6,9-cis-3,4-epoxy-nonadecadiene increases zinc bioavailability and availability of zinc boosts the immune system in the body. PENTADECANOIC ACID stops uric acid formation and uric acid is one of the major problems with non-vegetarian food habit people. Pentadecane is antibacterial, anti-inflammatory, anticancer; Dichloroacetic acid, tridec-2-ynyl ester and hexadecanoic acid, ethyl ester also inhibit uric acid formation; Hexadecanoic acid is hypocholesterolemic, and antioxidant, (2E)(7R,11R)-3,7,11,15-tetramethylhexadec-2-en-1-ol is Endoanesthetic, endocrinoprotective, gamma-Tocopherol is anticancer, anti-inflammatory, antioxidant, antitumor, cardioprotective, hypocholesterolemic, natriuretic. Presence of adequate amount of potassium, magnesium, iron, Carbohydrate, Crude protein and Crude fibre is important for a healthy diet. Minerals and Proximate compositions in a food are vital for the proper growth and development of a healthy body and secondary metabolites included in diet act as nutraceuticals thus help in fighting various health problems. The findings of Keawsa-ard *et al.*, [16]: also support the results and findings of present study in which the essential oil of *Solanum spirale* Roxb. unripe fruits is reported to exhibit anticancer activities against MCF-7 (breast cancer) and NCI-H187 (small cell lung cancer), antituberculosis activity against *Mycobacterium tuberculosis* H37Ra and also showed moderately antibacterial activity against *Escherichia coli* and *Staphylococcus aureus*. Likewise, Sukanya *et al.*, [17] also

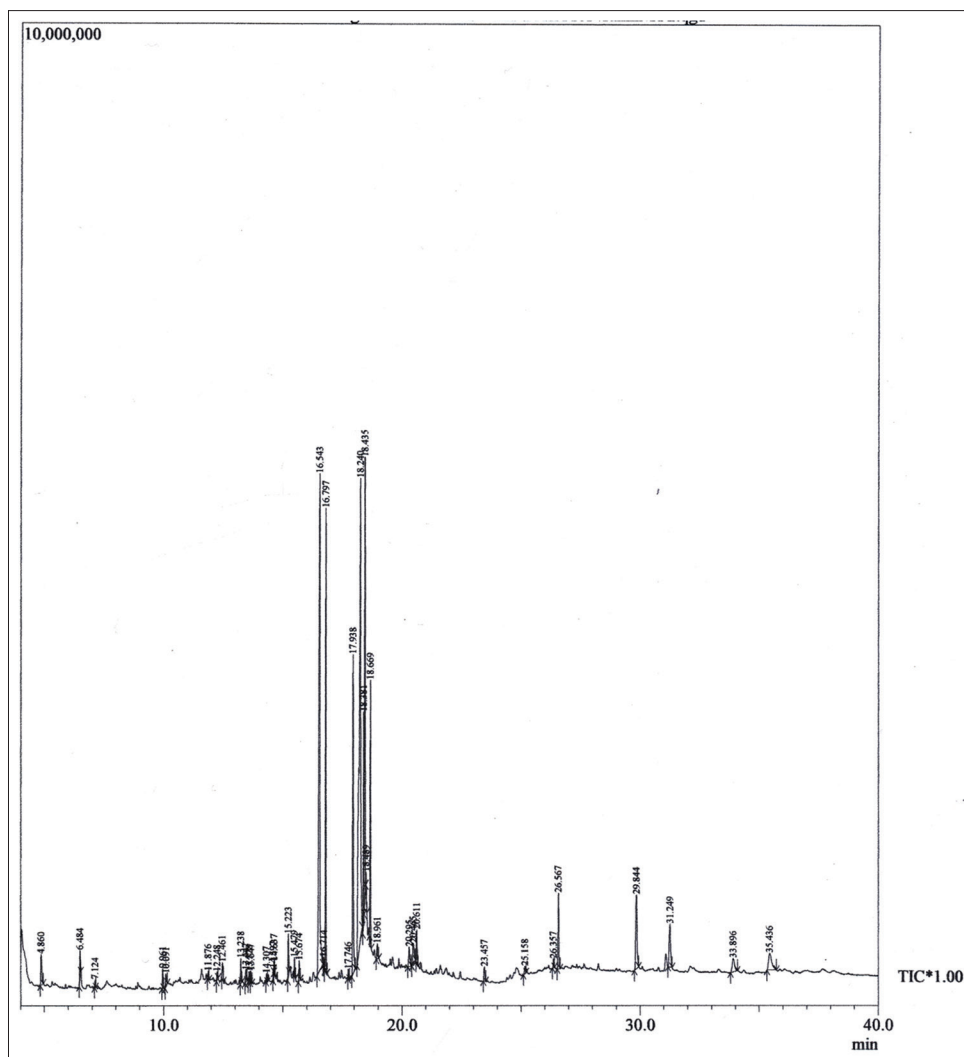


Figure 1: Chromatogram of ethanol extract *Solanum spirale* shoot.

reported the *Solanum spirale* leaf essential oil also reported that showed significant antibacterial activity against both Gram-negative *Escherichia coli* and Gram-positive *Staphylococcus aureus* and also showed significant cytotoxicity against KB (oral cancer), MCF-7 (breast cancer) and NCI-H187 (small cell lung cancer) and recently Payum *et. al.*, [18] have studied the Pharmacognostic characteristic of *Solanum spirale* shoot. In conclusion, *Solanum spirale* is a nutraceutical plant that provides energy, minerals including various useful phytochemicals. Presence of anti stroke and cardioprotective compounds validate the traditional knowledge and practices. The present study suggests the need of further studies on this nutraceutical plant.

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