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Research Article

STUDY ON GAS CHROMATOGRAPHY AND MASS SPECTROSCOPY ANALYSIS, ANTIOXIDANT, AND NUTRITIVE PROPERTIES OF *AERVA LANATA* (L.) JUSS. COLLECTED FROM SELECTED REGIONS OF SHIMOGA, KARNATAKA, INDIA

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ABSTRACT

Objective: *Aerva lanata* (L.) Juss., the plant ethanolic extract was subjected to gas chromatography and mass spectroscopy, antioxidant, and nutritive experiment using standard methods.

Methods: Antioxidant experiment is done using 2, 2-diphenyl-1-picrylhydrazyl, 2, 2-azobis-3-ethylbenthiozoline-6-sulfonic acid, superoxide radical scavenging, hydroxy radical scavenging, and metal-chelating assays. Nutritive value is performed by double acid digestion followed by atomic absorption spectroscopy.

Results: Antioxidant experiment revealed that *A. lanata* (L.) Juss. the plant ethanolic extract has good medicinal compounds exhibits excellent antioxidant activity in all tested experiments, but comparably less with the standards used. From nutritive value experiment, it is revealed that *A. lanata* (L.) Juss the plant has high iron content with rich macro- and micro-nutrients.

Conclusion: *A. lanata* (L.) Juss. could be exploited as a valuable source of antioxidant agent enriching with nutrients.

Keywords: *Aerva lanata* (L.) Juss., Antioxidant activity, Gas chromatography and mass spectroscopy, Nutritive properties.

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INTRODUCTION

Aerva lanata (L.) Juss. is a weed plant, distributed throughout India, commonly known as weed plant but has many traditional medicinal used.

Botanical description

Herb, erect, or prostate with a long taproot, branched from near the base; branches many, pubescent or woolly-tomentose, striate.

Leaves alternate, 2–2 cm × 1–1.6 cm on the main stem, 6–10 mm × 5–6 mm on the branched, elliptic or obovate, or suborbicular, obtuse or acute, entire, pubescent above, more or less white with cottony hairs beneath; petioles 3–6 mm long, often obscure.

Flowers greenish-white, very small, sessile, often bisexual, in small, dense subsessile axillary heads or spikes 6–13 mm long often closely crowded and forming globules clusters; bracteoles 1.25 mm, long, membranous, broadly ovate, concave, apiculate, perianth 1.5–1.25 mm long; sepals oblong, obtuse, sometimes articulate, silky-hairy on the back. Utricle broadly ovoid, acute, stigmas two, seed 0.85 mm in diameter, smooth and polished, black (Gamble*: A. lanata* (L.) Juss. Vol - I, 1883).

Common name

Mountain knotgrass; Hindi - Chaaya; Gorakbudi; Marathi - Kapurmadhuri; Tamil - ciras-pulai; Malayalam - Cherula; Kannada - bili hindi soppu; Telugu - pindidonda; and Sanskrit - asmahabodhaha.

Synonyms

Achyranthes lanata Roxb.; *A. elegans* Moq.; and *Illecebrum lanatum.*

Taxonomic hierarchy

Kingdom: Plantae
Subkingdom: Viridiplantae
Infrakingdom: Streptophyta
Superdivision: Embryophyta
Division: Tracheophyta
Subdivision: Spermatophytina
Class: Magnoliopsida
Superorder: Caryophyllanae
Order: Caryophyllales
Family: Amaranthaceae
Genus: *Aerva*
Species: *Lanata* (L.) Juss.

A. lanata (L.) Juss. traditionally used as painkiller in the treatment of headache and for cough, cutaneous infections, in white urine, diarrhea, cholera, dysentery, in kidney stone treatment, anthelmintic, strangury (slow to be and painful discharge of urine), headache, demulcent, anti-inflammatory, diuretic, hepatoprotective, hypoglycemic, antidiabetic, antiparasitic, antimicrobial, antiasthmatic, antifertility, hypolipidemic, antidiuretic, and nephroprotective property also used in the treatment of infections, cough, antidote, emollient, and skin infections [1-4].

Scientifically, *A. lanata* (L.) Juss. extracts proved for many pharmacological properties such as antinephrolithiasis, antiurolithiatic, diuretic, demulcent, anthelmintic hepatoprotective, anticancer, anti-inflammatory, hypoglycemic, antidiabetic, antiparasitic, hepoprotective, antiurolithiasis, antiasthmatic, antifertility, hypolipidemic, immunomodulatory, antioxidant, cytotoxic, immunomodulatory, and antimicrobial used in anti-HIV treatment and treatment in urinary complaints [5-18].

METHODS

Plant collection and authentication

The plant materials were collected Shimoga District, Karnataka, in January 2018. The plant was identified and authenticated by Dr. Sathish, Kuvempu University, and voucher specimen was conserved under the reference number KU/BD/VPY/001 (Fig. 2).

Plant preparation and extraction

The plant samples were dried in shade for 20–25 days, mechanically powdered, and subjected to Soxhlet extraction using ethanol. The crude extracts were collected in air-tight plastic containers and stored in cool condition (Fig. 1).

Preliminary phytochemical screening

Soxhlet extracted solvent crude extracts were screened for the presence of tannins, alkaloids, saponins, glycosides, flavonoids, steroids/sterols, and phenols using standard methods [19].

Gas chromatography and mass spectroscopy (GC–MS) analysis

Plant extracts were subjected to GC–MS obtained spectra were analyzed. GC model: Thermo Trace GC Ultra, MS model: Thermo DSQ II, Ionization: Electron impact ionisation (EI), chemical ionisation (CI), mass range: 1–1074 m/z. The oven temperature was kept at 70–80°C and was automated to reach 240–260°C at a rate of 8°C/min. Mass range was m/z 50–650. The total running time was completed in 50 min. The chromatogram obtained from GC was then analyzed in MS to get the mass of all fractions. The identification of phytoconstituents was accomplished by calculating retention time and mass spectra of unknown peaks were measured by comparing with Wiley 9 GC–MS library [20].

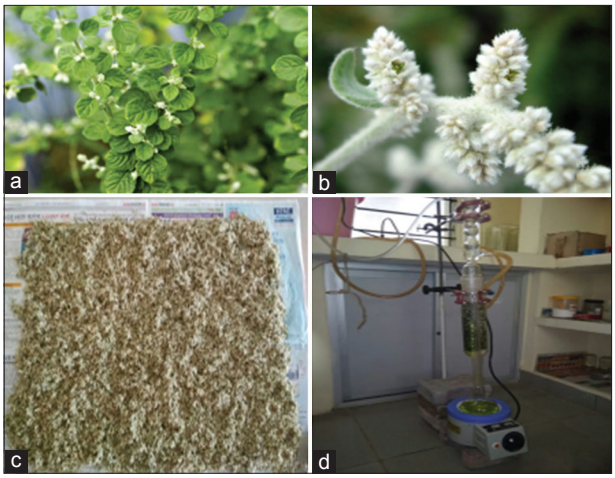

Fig. 1: *Aerva lanata* (L.) Juss. (a) aerial part, (b) inflorescence, (c) dried grinded sample, (d) Soxhlet extraction

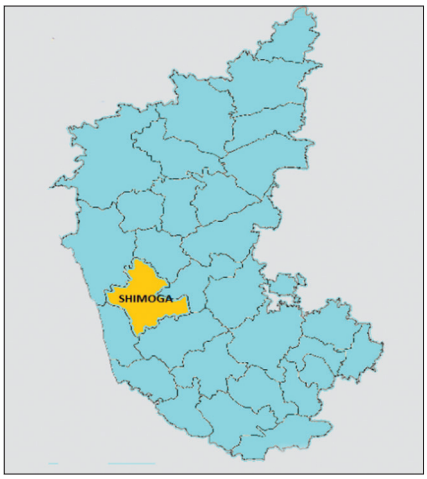

Fig. 2: Location where plants were collected

In vitro Antioxidant activity

Scavenging of 2, 2-diphenyl-1-picrylhydrazyl (DPPH) radicals

Stable radical DPPH[21] in methanol was used as a substrate to evaluate antioxidant activity. The method is based on the reduction of DPPH radical in the presence of a hydrogen donating antioxidants, leading to the formation of a non-radical form DPPH-H by the reaction. DPPH in its radical form has an absorption peak at 515 nm which disappeared on the reduction of antioxidant compounds. Absorbance was measured 20 min after the reaction was started.

Radical scavenging activity was calculated using the following formula:

$$Percentage\,inhibition = \frac{Absorbance\,of\,control - Absorbance\,of\,test}{Absorbance\,of\,control} \times 100 \quad (1)$$

Half maximal inhibitory concentration (IC50) value was calculated using the following formula:

$$Percentage\,inhibition = \frac{Absorbance\,of\,control - Absorbance\,of\,test}{Absorbance\,of\,control} \times 100 \quad (2)$$

Scavenging of 2, 2-azobis-3-ethylbenthiozoline-6-sulfonic acid (ABTS) radicals

The principle involved in ABTS radical scavenging activity[21] is oxidation of ABTS to its cation radicals by ferryl myoglobin formed in the reaction of H_2O_2 and metmyoglobin. Briefly, the stock solutions of 500 µM ABTS diammonium salt, 400 µM myoglobin (MbIII), 740 µM potassium ferricyanide, and 450 µM H_2O_2 were prepared in PBS (pH 7.4). Metmyoglobin was prepared by mixing equal volumes of myoglobin and potassium ferricyanide solutions. The reaction mixture (2 mL) contained ABTS (150 µM), MbIII (2.25 µM), and varying concentrations of extracts in PBS. The reaction was initiated by adding 75 µM H_2O_2 and oxidation reaction was monitored at 734 nm.

Elemental composition of *A. lanata* (L.) Juss. aerial parts

The microelements, calcium, magnesium, zinc, copper, manganese, lead, and cadmium were analyzed by atomic absorption spectra GBC 932 AA/AAS. For atomic absorption spectra, plant samples were predigested with nitric acid (HNO_3) and HCl in the ratio of 1:3 for 1–4 h depending on the plant sample. Then, the sample is kept over hot water bath (95°C) for 4–5 h till the sample completely dissolved [22,23].

RESULTS

Extracts yield of *A. lanata* (L.) Juss. aerial part ethanolic extract

Soxhlet extraction of *A. lanata* (L.) Juss. aerial part (700 g) with ethanol gives 35.12 g [Table 1].

Preliminary qualitative phytochemical analysis of *A. lanata* (L.) Juss. aerial part ethanolic extract

The preliminary phytochemical analysis of extracts was given in Table 2. The preliminary phytochemical analysis of ethanolic extract gives positive result for flavonoids, steroids, glycosides, and phenols.

Table 1: Extracts yield of *Aerva lanata* (L.) Juss. aerial part ethanolic extract

S No.	Plant part used	Sample used in grams	Solvent used	Extract yield in grams
1	*Aerva lanata* (L.) Juss. aerial part	700 g	Ethanol	35.12 g

Table 2: Preliminary qualitative phytochemical analysis of *Aerva lanata* (L.) Juss. aerial part ethanolic extract

S. No.	Secondary metabolites	Name of the test	Ethanolic extract
1	Alkaloids	Mayer's test	-
		Wagner's test	-
2	Saponins	Foam test	-
3	Tannins	Ferric chloride test	-
		Gelatin test	+
		Shenoda test	+
4	Flavonoids	Zinc HCl reduction test	+
		Alkaline reagent test	+
		Lead acetate test	+
		Ferric chloride test	
5	Steroids	Salkowski test	-
6	Glycosides	Legal's test	+
		Brown water test	+
		Keller–Kiliani test	+
7	Phenols	Ellagic acid test	+
8	Sterols	Liebermann Burchard test	-
9	Terpenoids	Salkowski's test	-

+Present, –Absent

Quantitative GC–MS analysis of *A. lanata* (L.) Juss. aerial part ethanolic extract

We took only ethanolic extract of *A. lanata* (L.) Juss. for GC–MS analysis (Table 3 and Figs. 3 and 4).

GC–MS analysis of *A. lanata* (L.) Juss. aerial part ethanolic extract confirms the presence of 76 compounds, of these 36 compounds were unknown and 40 compounds were known for its medicinal properties, most of them were flavoring agents 13 in numbers, followed by 10 food additive, eight compounds were antioxidant, five compounds have antimicrobial properties, four compounds were anticancer, and rest of them were diuretic, laxative, antineoplastic agent, antiallergic agents, antiseborrheics, insect pheromones, anti-inflammatory, adhesives and sealant chemical, agricultural chemicals (non-pesticidal), finishing agents, lubricants and lubricant additives, surface active agents, anticholinesterase, nasal decongestant, appetite suppressant, anticholesteremic, etc., (Figs 3 and 4; Table 3).

Majorpercentageofcompoundiscis,cis,cis-7,10,13-hexadecatrienal(17.81%) followed by.gamma.-sitosterol (11.45%) and pentadecanoic acid (10.09%) and the least percentage is 3,7,11,15-tetramethyl-2-hexadecen-1-ol (0.08%).

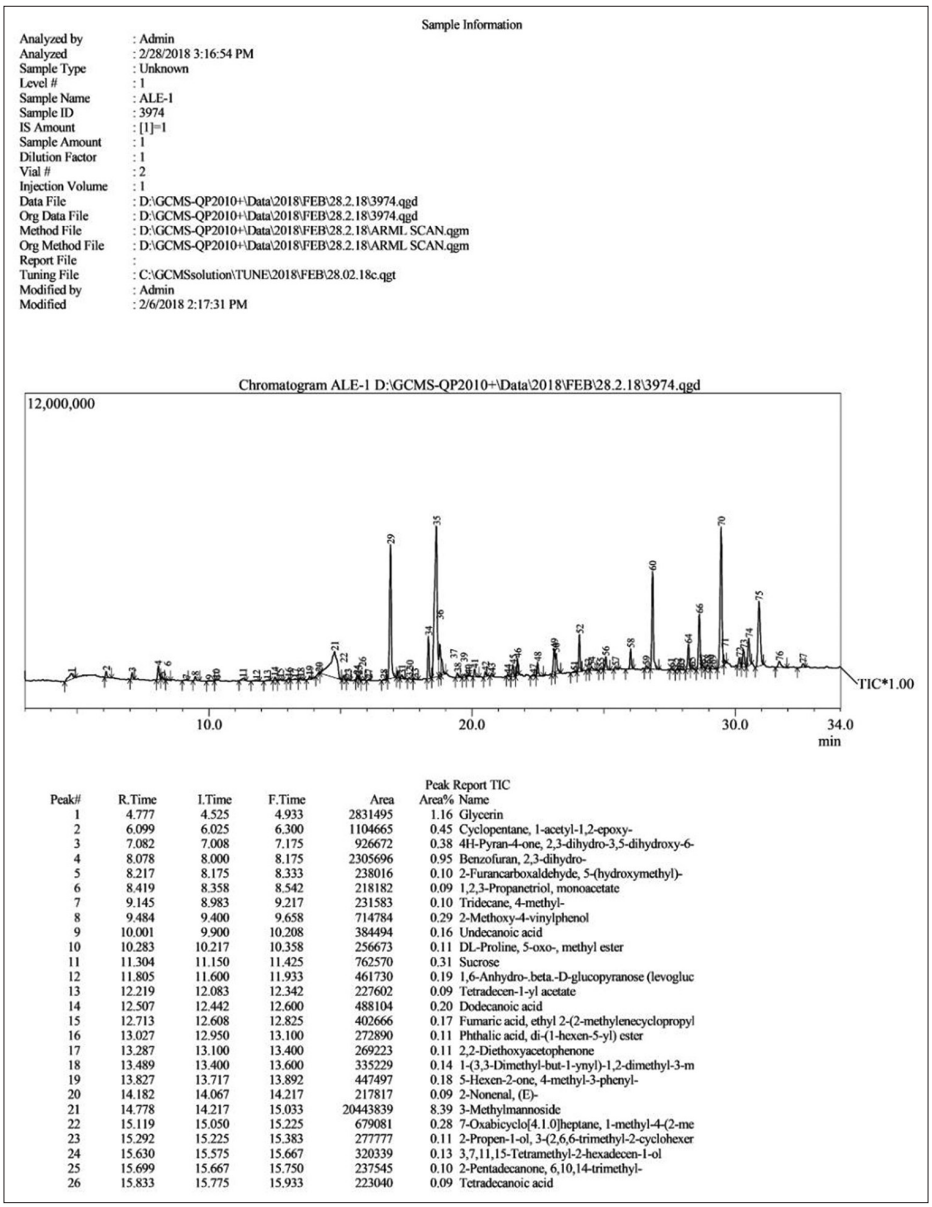

Fig. 3: Gas chromatography and mass spectroscopy analysis of *Aerva lanata* (L.) Juss. aerial part ethanolic extract

Table 3: GC–MS analysis of *Aerva lanata* (L.) Juss. aerial part ethanolic extract

S. No.	Phytocompounds in *Aerva lanata* (L.) Juss.	Percentage	Properties
1	Glycerin	1.16	Flavoring agent, food additive, diuretic, and laxative effects [24]
2	Cyclopentane, 1-acetyl-1,2-epoxy-	0.45	Unknown
3	4H-pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-	0.38	Mutagen, antimicrobial, anti-inflammatory, and antioxidant capacity [25-27]
4	Benzofuran, 2,3-dihydro-	0.95	Unknown
5	2-furancarboxaldehyde, 5-(hydroxymethyl)-	0.10	Antimicrobial, preservative, flavoring agents, food additives, component of onion, tomatoes, tobacco oil, etc., [28,29]
6	1,2,3-propanetriol, monoacetate	0.09	Food additives [30]
7	Tridecane, 4-methyl-	0.10	Antiandrogenic agent, antineoplastic agent, antiallergic agents, antiseborrheics [31]
8	2-methoxy-4-vinylphenol	0.29	Can induce cell cycle arrest, antibacterial, anti-inflammatory, antioxidant, flavoring agent, also acts as insect pheromones [32]
9	Undecanoic acid	0.16	Antifungal agent, to treat ringworm and athlete's foot, food additives, flavoring agent [33]
10	DL-proline, 5-oxo-, methyl ester	0.11	Unknown
11	Sucrose	0.31	Sweetener in foods and soft drinks, in the manufacture of syrups, in invert sugar, confectionery, preserves and jams, demulcent, pharmaceutical products, and caramel. Sucrose is also a chemical intermediate for detergents, emulsifying agents, and other sucrose derivatives. Sucrose is widespread in seeds, leaves, fruits, flowers, and roots of plants, where it functions as an energy store for metabolism and as a carbon source for biosynthesis. The annual world production of sucrose is in excess of 90 million tons mainly from the juice of sugar cane (20%) and sugar beet (17%). In addition to its use as a sweeter, sucrose is used in food products as a preservative, antioxidant, moisture control agent, stabilizer, and thickening agent [34]
12	1,6-anhydro-.beta.-D-glucopyranose (levoglucosan)	0.19	Formed by pyrolysis of carbohydrates such as starch and cellulose. As a result, levoglucosan is often used as a chemical tracer for biomass burning in atmospheric chemistry studies, used in the production of bioethanol [35]
13	Tetradecen-1-yl acetate	0.09	Unknown
14	Dodecanoic acid	0.20	Found naturally in various plant and animal fats and oils, and is a major component of coconut oil and palm kernel oil. Have antimicrobial properties used in many soaps and shampoos [36]
15	Fumaric acid, ethyl 2-(2-methylenecyclopropyl) propyl	0.17	Unknown
16	Phthalic acid, di-(1-hexen-5-yl) ester	0.11	Unknown
17	2,2-diethoxyacetophenone	0.11	Unknown
18	1-(3,3-dimethyl-but-1-ynyl)-1,2-dimethyl-3-methylene-cyclopr	0.14	Unknown
19	5-hexen-2-one, 4-methyl-3-phenyl-	0.18	Unknown
20	2-nonenal, (E)-	0.09	Flavoring agents, food additive, agonists of the AR signaling pathway [37]
21	3-methylmannoside	8.39	Unknown
22	7-oxabicyclo[4.1.0]heptane, 1-methyl-4-(2-methyloxiranyl)-	0.28	Moderately toxic [38]
23	2-propen-1-ol, 3-(2,6,6-trimethyl-2-cyclohexen	0.11	Unknown
24	3,7,11,15-tetramethyl-2-hexadecen-1-ol	0.13	A precursor for the manufacture of synthetic forms of Vitamin E and Vitamin K1, flavoring agents [39]
25	2-pentadecanone, 6,10,14-trimethyl-	0.10	Flavoring agents [40]
26	3,7,11,15-tetramethyl-2-hexadecen-1-ol	0.08	Phytol is an acyclic diterpene alcohol and a constituent of chlorophyll. Phytol is commonly used as a precursor for the manufacture of synthetic forms of Vitamin E and Vitamin K1 [39]
27	Methyl-Z, Z-3,13-octadecadieno	0.10	Unknown
28	Pentadecanoic acid	10.09	Adhesives and sealant chemical, agricultural chemicals (non-pesticidal), finishing agents, lubricants and lubricant additives, surface active agents [41]
29	Hexadecanoic acid, ethyl ester	0.38	Flavoring agents [42]
30	1,3-benzenedimethanol, 2-hydroxy-5-methyl-	0.26	Unknown

(Contd...)

Table 1: (*Continued*)

S. No.	Phytocompounds in *Aerva lanata* (L.) Juss.	Percentage	Properties
31	Oleic acid	0.14	Component in many foods, in the form of its triglycerides. It is a component of the normal human diet as a part of animal fats and vegetable oils. Oleic acid as its sodium salt is a major component of soap as an emulsifying agent. It is also used as an emollient. Small amounts of oleic acid are used as an excipient in pharmaceuticals, and it is used as an emulsifying or solubilizing agent in aerosol products [43]
32	Tetradecanoic acid	0.18	Occurring in most animal and vegetable fats, used to synthesize flavor and as an ingredient in soaps and cosmetics [44]
33	Phytol	2.40	A precursor for the manufacture of synthetic forms of Vitamin E and Vitamin K1, flavoring agents[45]
34	cis, cis, cis-7,10,13-hexadecatrienal	17.81	Unknown
35	9,12-octadecadienoic acid (Z, Z)-	2.21	Treatment in the prevention of preeclampsia, flavoring agents, antioxidant, anticholinesterase, and antibacterial activities [46]
36	9,12,15-octadecatrienoic acid, ethyl ester, (Z, Z, Z)-	0.96	Unknown
37	Ambrettolide	0.27	Flavoring agents in dairy products [47]
38	Benzyl (9Z)-9-octadecenoate	0.23	Unknown
39	Hexadecanal, 2-methyl-	0.09	Unknown
40	2,7-diazido-9H-fluorene	0.53	Unknown
41	2H-pyran-2-one, tetrahydro-6-tridecyl-	0.30	Flavoring agent[48]
42	Sulfurous acid, octadecyl 2-propyl ester	0.18	Unknown
43	Benzedrex	0.09	Is a nasal decongestant, appetite suppressant, and psychostimulant medication. It is an analog of methamphetamine that is used medicinally for relief of congestion due to colds, allergies, and allergic rhinitis and recreationally for its euphoric effects. Used to treat acute nasal congestion related to common cold, allergies, and hay fever [49]
44	Tritetracontane	0.81	Unknown
45	Hexadecanoic acid, 2-hydroxy-1-(hydroxymethyl)	0.89	Unknown
46	Sulfurous acid, octadecyl 2-propyl ester	0.14	Unknown
47	3-methyl-1-oxo-2,3-dihydro-1H-pyrazolo[4,3-c][1,10] phenanthroline	0.97	Unknown
48	(R)-(-)-14-methyl-8-hexadecyn-1-ol	1.77	Unknown
49	Methyl (Z)-5,11,14,17-eicosatetraenoate	1.51	Unknown
50	Z, Z-8,10-hexadecadien-1-ol	0.10	Unknown
51	2,6,10,14,18,22-tetracosahexaene, 2,6,10,15,19,23-hexamethyl-, (all-E)-	2.08	Squalene is originally obtained from shark liver oil. It is a natural 30-carbon isoprenoid compound and intermediate metabolite in the synthesis of cholesterol. It is not susceptible to lipid peroxidation and provides skin protection. It is ubiquitously distributed in human tissues where it is transported in serum generally in association with very low-density lipoproteins. Squalene is investigated as an adjunctive cancer therapy [50]
52	17-pentatriacontene	0.12	Unknown
53	1-hexacosanol	0.37	It occurs naturally in the epicuticular wax and plant cuticle of many plant species [51]
54	1,6,10,14,18,22-tetracosahexaen-3-ol, 2,6,10,15,19,23-hexamethyl-, (all-E)-	0.14	Unknown
55	2H-1-benzopyran-6-ol, 3,4-dihydro-2,8-dimethyl-2-(4,8,12-trimethyltridecyl)-, [2R-[2R*(4R*,8R*)]]-	0.82	Vitamin E component, food additive found in certain nuts and seeds, with potential antioxidant activity [52]
56	Tetracosapentaene, 2,6,10,15,19,23-hexamethyl-	0.20	Unknown
57	.Gamma.-tocopherol	1.33	Vitamin E component, food additive found in certain nuts and seeds, with potential antioxidant activity [53]
58	17-pentatriacontene	0.17	Unknown
59	Vitamin E	6.90	Vitamin E, known for its antioxidant activities, is protective against cardiovascular disease and some forms of cancer and has also demonstrated immune-enhancing effects. It may be of limited benefit in some with asthma and rheumatoid arthritis. It may be helpful in some neurological diseases including

(Contd...)

Table 1: (*Continued*)

S. No.	Phytocompounds in *Aerva lanata* (L.) Juss.	Percentage	Properties
			Alzheimer's, some eye disorders including cataracts, and diabetes, and premenstrual syndrome. It may also help protect skin from ultraviolet irradiation although claims that it reverses skin aging, enhances male fertility, and exercise performance is poorly supported. It may help relieve some muscle cramps [54]
60	delta. 8-cholesten-3.beta.-ol	0.12	Unknown
61	Ergost-8-en-3-ol, 14-methyl-, (3.beta., 5.alpha.)-	0.12	Unknown
62	22,23-dibromostigmasterol acetate	0.09	Unknown
63	Campesterol	1.86	Naturally occurs in many vegetables, fruits, nuts, and seeds, anticholesteremic [55]
64	9,19-cycloergost-24 (28)-en-3-ol, 4,14-dimethyl-, acetate, (3.beta.,4.alpha.,5.alpha.)-	0.11	Unknown
65	Stigmasterol	3.91	Inhibits certain cancers, food additive, cytotoxicity, antioxidant, hypoglycemic, and thyroid-inhibiting properties [56]
66	Ergost-25-ene-3,5,6,12-tetrol, (3.beta.,5.alpha.,6.beta.,12.beta.)-	0.16	Unknown
67	7-ergosteno	0.30	Unknown
68	Lanosterol	0.41	Intermediate of cholesterol biosynthesis in animals and fungus [57]
69	Gamma.-sitosterol	11.45	Hypolipidemic agents isolated from the Indian marine red alga *Gracilaria edulis*, the sponge Veronica aerophoba and the Kenyan Marine Green. Macroalga *Halimeda macroloba* [58]
70	Stigmastanol	0.22	Inhibits biosynthesis of cholesterol, anticholesteremic agents [59]
71	4,4,6a, 6b, 8a, 11,11,14b-octamethyl-1,4,4a, 5,6,6a, 6b, 7,8,8a, 9,10,11,12,12a, 14,14a	0.60	Unknown
72	Stigmast-7-en-3-ol, (3.beta.,5.alpha.,24S)-	1.32	Component of ethanolic extract of *Chenopodium formosanum* [60]
73	Lupeol	2.19	Naturally found in fruits and vegetables, has anti-inflammatory agents, analgesic, anticancer activity, cytotoxicity [61,62]
74	Lup-20 (29)-en-3.beta.-ol	7.02	Unknown
75	3.alpha.,7.beta.-dihydroxy-5.beta.,6.beta.-epoxycholestane	0.76	Used in the drug manufacturing [63]
76	Acetic acid, 3-hydroxy-6-isopropenyl-4,8a-dimethyl-1,2,3,5,6,7,8,8a	0.35	Unknown

AR: Androgen receptor, GC–MS: Gas chromatography and mass spectroscopy

***In vitro* antioxidant properties of *A. lanata* (L.) Juss. aerial part ethanolic extract**

A. lanata (L.) *Juss.* aerial part ethanolic extract subjected to different antioxidant experiments such as DPPH and ABTS. The experiments were triplicated and values were expressed in terms of mean ± standard error of mean.

DPPH radical scavenging activity of A. lanata (L.) Juss. aerial part ethanolic extract

In DPPH radical scavenging activity, *A. lanata* (L.) Juss. aerial part ethanolic extract showed dose-dependent radical scavenging activity in all tested concentrations. IC_{50} value of the ethanolic crude extract (129.93 ± 7.07) is not equal to the value of standard ascorbic acid (39.48 ± 0.02) used, showed moderate antioxidant properties (Table 4 and Fig. 5).

ABTS radical scavenging activity of *A. lanata* (L.) Juss. aerial part ethanolic extract

In ABTS radical scavenging activity of *A. lanata* (L.) Juss. aerial part ethanolic extract showed dose-dependent antioxidant activity in all tested concentrations. IC_{50} value of the ethanolic crude extract (84.37 ± 2.68) is not equal to the value of standard butylated hydroxyl anisole (66.92 ± 0.36) used, showed moderate antioxidant properties (Table 5 and Fig. 6).

Elemental composition of sample

A. lanata (L.) Juss. aerial part sample subjected for nutrient analysis through atomic absorption spectroscopy. The results were found to be satisfying with sufficient quantity of macronutrients such as nitrogen (3.08 ± 0.34), phosphorus (0.15 ± 0.14), potassium (2.49 ± 0.32), calcium, and magnesium showed the nil percentage (Table 6 and Fig. 7). Similarly, micronutrients like iron (404.00 ± 0.34), manganese (39.20 ± 0.52), zinc (27.40 ± 0.62) and copper (7.85 ± 0.73) were confirmed in appreciable percentage. The values was obtained were expressed in terms of ppm (parts per million) (Table 7 and Fig. 8). In all the nutrient components of *A. lanata* (L.) Juss. aerial part sample, the iron (404.00 ± 0.34) was found to be highest, which is essential micronutrient mainly help in the treatment of anemic patients having the deficiency of iron in the form of ferrous ion. In developing countries, iron deficiency is the common factor affects the growth of devolving children. Ferrous ions also help in the hemoglobin formation which essential for human beings.

DISCUSSION

Soxhlet extraction of *A. lanata* (L.) Juss. aerial part

Soxhlet extraction is a common procedure to extract phytoconstituents which is essential to humankind. The aerial part sample (700 g) of *A. lanata* (L.) Juss. aerial part yields maximum percentage of extract in ethanolic extract (35.12 g) so it is revealed that the plant aerial part sample is having more alcohol-soluble extractive which is more essential in extraction of good phytoconstituent [Table 1].

Preliminary phytochemical analysis of *A. lanata* (L.) Juss. aerial part ethanolic extract

The preliminary phytochemical analysis of *A. lanata* (L.) Juss. aerial part extracts also revealed the presence of more phytoconstituent in the ethanolic extracts such as alkaloids, saponins, flavonoids, steroids, glycosides, phenols, and sterols. Hence, we took only ethanolic extract for GC–MS analysis for confirmation of different constituents [Table 2].

Peak#	R.Time	I.Time	F.Time	Area	Area%	Name
27	16.073	15.992	16.142	203037	0.08	3,7,11,15-Tetramethyl-2-hexadecen-1-ol
28	16.655	16.567	16.725	233658	0.10	Methyl-Z,Z-3,13-octadecadieno
29	16.904	16.792	17.117	24589707	10.09	Pentadecanoic acid
30	17.173	17.117	17.258	914667	0.38	Hexadecanoic acid, ethyl ester
31	17.329	17.267	17.442	629468	0.26	1,3-Benzenedimethanol, 2-hydroxy-5-methyl-
32	17.616	17.542	17.750	344740	0.14	Oleic Acid
33	17.820	17.758	17.958	207252	0.09	Tetradecanoic acid
34	18.344	18.275	18.458	5844781	2.40	Phytol
35	18.645	18.467	18.733	43402771	17.81	cis,cis,cis-7,10,13-Hexadecatrienal
36	18.776	18.733	18.825	5375974	2.21	9,12-Octadecadienoic acid (Z,Z)-
37	18.841	18.825	19.000	2335216	0.96	9,12,15-Octadecatrienoic acid, ethyl ester, (Z,Z
38	19.448	19.367	19.558	660341	0.27	Ambrettolide
39	19.692	19.617	19.783	559985	0.23	Benzyl (9Z)-9-octadecenoate
40	19.852	19.792	19.892	231522	0.09	Hexadecanal, 2-methyl-
41	20.095	20.033	20.275	1281976	0.53	2,7-Diazido-9H-fluorene
42	20.510	20.433	20.575	719543	0.30	2H-Pyran-2-one, tetrahydro-6-tridecyl-
43	20.749	20.675	20.817	445224	0.18	Sulfurous acid, octadecyl 2-propyl ester
44	21.356	21.308	21.417	228735	0.09	Benzedrex
45	21.551	21.475	21.650	1981692	0.81	Tritetracontane
46	21.725	21.658	21.783	2171889	0.89	Hexadecanoic acid, 2-hydroxy-1-(hydroxymeth
47	22.329	22.217	22.417	340876	0.14	Sulfurous acid, octadecyl 2-propyl ester
48	22.492	22.417	22.708	2371418	0.97	3-Methyl-1-oxo-2,3-dihydro-1H-pyrazolo[4,3-c
49	23.123	23.025	23.158	4308631	1.77	(R)-(-)-14-Methyl-8-hexadecyn-1-ol
50	23.192	23.158	23.383	3683411	1.51	Methyl (Z)-5,11,14,17-eicosatetraenoate
51	23.890	23.742	23.950	250483	0.10	Z,Z-8,10-Hexadecadien-1-ol
52	24.081	23.992	24.175	5068931	2.08	2,6,10,14,18,22-Tetracosahexaene, 2,6,10,15,1
53	24.403	24.325	24.458	288393	0.12	17-Pentatriacontene
54	24.542	24.458	24.742	912185	0.37	1-Hexacosanol
55	24.879	24.817	24.992	335534	0.14	1,6,10,14,18,22-Tetracosahexaen-3-ol, 2,6,10,1
56	25.078	24.992	25.250	2003159	0.82	2H-1-Benzopyran-6-ol, 3,4-dihydro-2,8-dimeth
57	25.443	25.375	25.558	484025	0.20	Tetracosapentaene, 2,6,10,15,19,23-hexamethy
58	26.024	25.842	26.133	3241768	1.33	.gamma.-Tocopherol
59	26.627	26.558	26.692	405448	0.17	17-Pentatriacontene
60	26.857	26.767	27.100	16821569	6.90	Vitamin E
61	27.574	27.500	27.650	298625	0.12	delta.8-Cholesten-3.beta.-ol
62	27.803	27.717	27.883	294478	0.12	Ergost-8-en-3-ol, 14-methyl-, (3.beta.,5.alpha.)
63	27.948	27.883	28.050	218684	0.09	22,23-Dibromostigmasterol acetate
64	28.222	28.117	28.308	4521557	1.86	Campesterol
65	28.359	28.308	28.467	275208	0.11	9,19-Cycloergost-24(28)-en-3-ol, 4,14-dimethy
66	28.633	28.525	28.725	9534848	3.91	Stigmasterol
67	28.773	28.725	28.842	401356	0.16	Ergost-25-ene-3,5,6,12-tetrol, (3.beta.,5.alpha.,
68	28.961	28.858	29.050	739146	0.30	7-Ergostenol
69	29.126	29.050	29.308	998720	0.41	Lanosterol
70	29.467	29.325	29.567	27904481	11.45	.gamma.-Sitosterol
71	29.606	29.567	29.658	541535	0.22	Stigmastanol
72	30.161	30.075	30.233	1470490	0.60	4,4,6a,6b,8a,11,11,14b-Octamethyl-1,4,4a,5,6,6
73	30.316	30.233	30.417	3209354	1.32	Stigmast-7-en-3-ol, (3.beta.,5.alpha.,24S)-
74	30.520	30.425	30.625	5339031	2.19	Lupeol
75	30.908	30.767	31.067	17111497	7.02	Lup-20(29)-en-3.beta.-ol
76	31.678	31.533	31.967	1851175	0.76	3.alpha.,7.beta.-Dihydroxy-5.beta.,6.beta.-epox
77	32.578	32.358	32.675	860419	0.35	Acetic acid, 3-hydroxy-6-isopropenyl-4,8a-dim
				243727822	100.00	

Fig. 4: Gas chromatography and mass spectroscopy analysis of *Aerva lanata* (L.) Juss. aerial part ethanolic extract

Table 4: DPPH radical scavenging activity of *Aerva lanata* (L.) Juss. aerial part ethanolic extract

Activity	Extracts	Concentration in µg/mL	Scavenging activity	IC_{50} value	Standard µg/mL (ascorbic acid)	IC_{50} value of standard ascorbic acid
DPPH activity	Ethanolic	25	17.33 ± 0.88	129.93 ± 7.07	76.23 ± 0.23	39.48 ± 0.02
		50	21.66 ± 0.66		82.32 ± 0.43	
		75	31.33 ± 0.33		113.11 ± 0.09	
		100	39.66 ± 0.66		134.54 ± 0.91	
		125	47.66 ± 2.18		156.43 ± 0.02	
		150	57 ± 1.15		176.65 ± 0.34	
		175	62.33 ± 0.88		189.41 ± 0.54	
		200	69.33 ± 0.33		210.87 ± 0.32	

IC_{50}: Half maximal inhibitory concentration, DPPH: 2, 2-diphenyl-1-picrylhydrazyl

Table 5: ABTS radical scavenging activity of *Aerva lanata* (L.) Juss. aerial part ethanolic extract

Activity	Extracts	Concentration in µg/mL	scavenging activity	IC_{50} value	Standard µg/mL (butylated hydroxyl anisole)	IC_{50} value of standard butylated hydroxyl anisole
ABTS activity	Ethanolic	50	7±0.57	382.43±1.34	47.34±0.32	66.92±0.36
		100	12.33±0.66		84.65±0.05	
		150	18.33±0.88		120.43±0.36	
		200	23±1.15		149.68±0.1	
		250	33.66±2.02		185.65±0.3	
		300	39.33±1.45		214.76±0.62	
		350	46.33±0.88		254.36±0.06	
		400	55.33±2.33		287.98±0.6	

IC_{50}: Half maximal inhibitory concentration, ABTS: 2, 2-azobis-3-ethylbenthiozoline-6-sulfonic acid

Table 6: Macronutrient of *Aerva lanata* (L.) Juss. aerial sample

S. No.	Samples	Macronutrients in percentage (mean±SD)				
		N	P	K	Ca	Mg
1	Arial part	3.08±0.34	0.15±0.14	2.49±0.32	-	-
	Occurrence	Medium	Medium	Medium	Medium	Medium

SD: Standard deviation

Table 7: Micronutrients of *Aerva lanata* (L.) Juss. aerial sample

S. No.	Samples	Micronutrients in ppm (mean±SD)			
		Fe	Mn	Zn	Cu
1	Arial part	404.00±0.34	39.20±0.52	27.40±0.62	7.85±0.73
	Occurrence	High	Medium	Medium	Medium

SD: Standard deviation

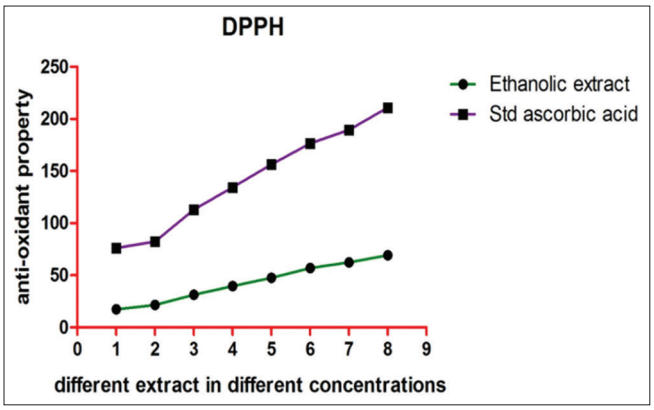

Fig. 5: 2, 2-diphenyl-1-picrylhydrazyl radical scavenging activity of *Aerva lanata* (L.) Juss. aerial part ethanolic extract

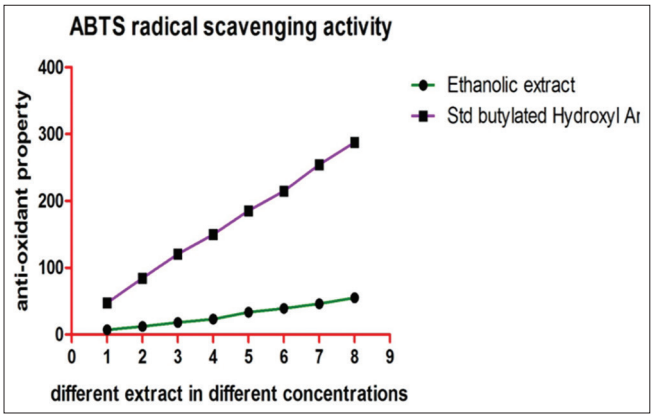

Fig. 6. 2, 2-azobis-3-ethylbenthiozoline-6-sulfonic acid radical scavenging activity of *Aerva lanata* (L.) Juss. aerial part ethanolic extract

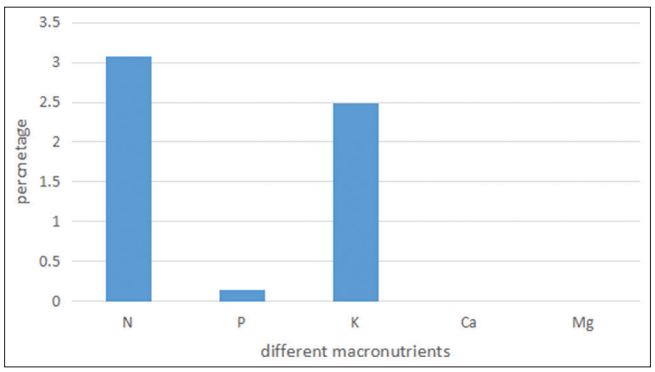

Fig. 7: Macronutrients of *Aerva lanata* (L.) Juss. aerial sample

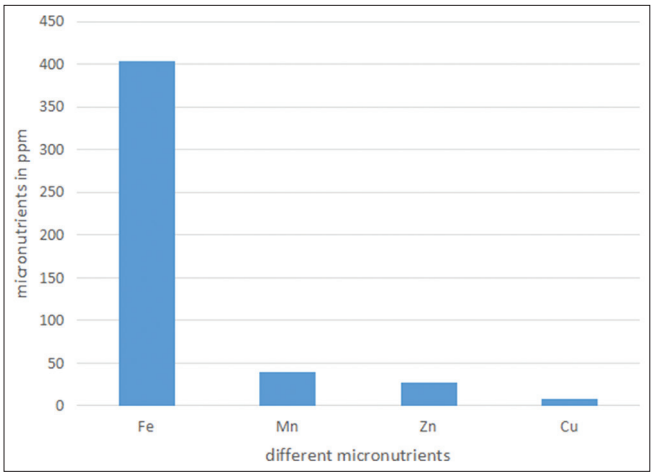

Fig. 8: Micronutrients of *Aerva lanata* (L.) Juss. aerial sample

GC–MS analysis of *A. lanata* (L.) Juss. aerial part ethanolic extract

GC–MS analysis of *A. lanata* (L.) Juss. aerial part ethanolic extract

was analyzed in the instrument GC Model: Thermo Trace GC Ultra, MS model: Thermo DSQ II, Ionization: EI, chemical ionization (CI), and mass range: 1–1074 m/z and obtained spectra were analyzed, revealed the presence of 76 compounds in that 36 compounds were unknown and 40 compounds were known for its medicinal properties (Figs 3 and 4; Table 3).

Major percentage of compound is cis, cis, cis-7,10,13-Hexadecatrienal (17.81%) unknown for its pharmacological properties, followed by.gamma.-sitosterol (11.45%) used as hypolipidemic agents, isolated from the Indian marine red alga *Gracilaria edulis*, the sponge *Veronica aerophoba* and the Kenyan marine green Macroalga *Halimeda* [58], pentadecanoic acid (10.09%) adhesives and sealant chemical, agricultural chemicals (non-pesticidal), finishing agents, lubricants and lubricant additives, and surface active agents [40], and the least percentage is 3,7,11,15-Tetramethyl-2-hexadecen-1-ol (0.08%) used as a precursor for the manufacture of synthetic forms of Vitamin E and Vitamin K1 [45].

13 compounds were known as flavoring agents used in food industries such as glycerin; 2-furancarboxaldehyde, 5-(hydroxymethyl)-; 2-methoxy-4-vinylphenol; undecanoic acid; 2-nonenal, (E)-; 3,7,11,15-tetramethyl-2-hexadecen-1-ol; 2-pentadecanone, 6,10,14-trimethyl-; hexadecanoic acid, ethyl ester; tetradecanoic acid; phytol; 9,12-octadecadienoic acid (Z, Z)-; ambrettolide; and 2H-pyran-2-one, tetrahydro-6-tridecyl-.

10 compounds were known as food additives, used in food and nutrient industries such as glycerin; 2-furancarboxaldehyde, 5-(hydroxymethyl)-; 1,2,3-propanetriol, monoacetate; undecanoic acid; sucrose; oleic acid; 2H-1-benzopyran-6-ol, 3,4-dihydro-2,8-dimethyl-2-(4,8,12-trimethyltridecyl)-, [2R-[2R*(4R*,8R*)]]-;.gamma.-tocopherol; stigmasterol; and 2-nonenal, (E)- 4H-pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-

Eight compounds were known for its antioxidant properties, namely 2-methoxy-4-vinylphenol; sucrose; 9,12-octadecadienoic acid (Z, Z)-; 2H-1-benzopyran-6-ol, 3,4-dihydro-2,8-dimethyl-2-(4,8,12-trimethyltridecyl)-, [2R-[2R*(4R*,8R*)]]-; .gamma.-tocopherol; Vitamin E; and stigmasterol.

Five compounds were known for its antimicrobial properties, namely 4H-pyran-4-one, 2,3-dihydro-3,5-dihydroxy-6-methyl-; 2-furancarboxaldehyde, 5-(hydroxymethyl)-; dodecanoic acid; undecanoic acid; and lanosterol.

Four compounds were known for its anticancer properties, namely 2-methoxy-4-vinylphenol; stigmasterol; Vitamin E; and lupeol.

Rest of them were diuretic, laxative, antineoplastic agent, antiallergic agents, antiseborrheics, insect pheromones, anti-inflammatory, adhesives and sealant chemical, agricultural chemicals (non-pesticidal), finishing agents, lubricants and lubricant additives, surface active agents, anticholinesterase, nasal decongestant, appetite suppressant, anticholesteremic, etc.

Capesterol is even though present in less quantity has many pharmacological properties such as antibacterial [64,65], antinociceptive [66], and anticarcinogenic [67], and identified in most of the plants such as *Syzygium aromaticum*[65] and *Chrysanthemum coronarium* [67].

***In vitro* antioxidant properties of *A. lanata* (L.) Juss. aerial part ethanolic extract**

In our study, it is revealed that *A. lanata* (L.) Juss. aerial part ethanolic extract showed appreciable antioxidant activity in all tested concentrations which is not comparable with the standards used. The antioxidant property of *A. lanata* (L.) Juss. aerial part ethanolic extract is may be due to the presence of eight compounds present in it, such as, 2-methoxy-4-vinylphenol; sucrose; 9,12-octadecadienoic acid (Z, Z)-; 2H-1-benzopyran-6-ol, 3,4-dihydro-2,8-dimethyl-2-(4,8,12-trimethyltridecyl)-, [2R-[2R*(4R*,8R*)]]-;.gamma.-tocopherol; Vitamin E; and stigmasterol (Tables 4 and 5 and Figs. 5 and 6).

Elemental composition of *A. lanata* (L.) Juss. aerial part

Macronutrient

Potassium (K) was found in normal level in *Aerva lananta* aerial parts. We need at least 4–5 g of potassium per day to perform normal work, maintain a normal blood pressure, work as an electrolyte in maintaining the body fluid, and maintain normal bone strength.

A. lanata (L.) Juss. aerial parts contain nitrogen (N) percentage in normal level. Nitrogen is a main component of DNA, RNA, and amino acids, an essential nutrient mainly required for protein synthesis and enzymes.

A. lanata (L.) Juss. aerial parts contain phosphorus (P) was found in normal level. In the human body, phosphorus has a vital role in the formation of bone and teeth, helps in the metabolism of carbohydrates and fats, helps in the synthesis of proteins in the human body, and triggers tissue repair mechanism (Table 6 and Fig. 7).

Micronutrient

In *A. lanata* (L.) Juss. aerial parts, the copper was found to be normal. Copper is essential for maintenance of brain health, antioxidant defense, the main component of neuron communication, and essential in maintenance of healthy skin.

In *A. lanata* (L.) Juss. aerial parts, the iron (Fe) was found to be high. The leaves have highest iron content. In human body, iron is an essential component of red blood cells, an important component of some proteins and enzymes, and also acts as enzyme cofactor, normal function of hemoglobin, and myoglobin.

In *A. lanata* (L.) Juss. aerial parts, manganese (Mn) was found to be in normal condition. In humans, manganese works as mettaloenzymes in the activation of enzyme-substrate reaction, also present in bone, cartilages, and proper functioning of thyroid.

In *A. lanata* (L.) Juss. aerial parts, zinc (Zn) was found to be in normal condition. In human body, zinc plays a key role in proper function of immune system by activation T-lymphocytes, proper neurophysiological function (Table 7 and Fig. 8).

CONCLUSION

After the present investigation, it can be concluded that the aerial part ethanolic extract of *A. lanata* (L.) Juss. can act as good antioxidant, as well as having good nutritive value. GC–MS analysis of ethanolic extract revealed the presence of 76 compounds in that 40 compounds were known for its medicinal properties, most of them were antimicrobial agents followed by food additive and flavoring agents, antioxidant, anticancer agents, antihypercholesterolemic compounds, anti-inflammatory agents, analgesic, etc.

The overall study on antioxidant, GC–MS, and elemental nutritive value reports that the plant species contain many active compounds which by their synergistic effect may reduce the inflammation and has an edible value with rich nutrients. Hence, it is finally concluded that the aerial part ethanolic extract of *A. lanata* (L.) Juss. can be explored for potential antioxidant and nutritive compounds.

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AUTHORS' CONTRIBUTION

Mr. Venkatesh Prasad Yadav and Ashwathanarayana R have collected the data, conducted the experiment and Mrs. Padmashree MS drafted the article.

CONFLICTS OF INTEREST

The authors declared that there are conflicts of interest.

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