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Research Article

PHYTOCHEMICALS SCREENING AND MINERALS STATUS OF *Mollugo cerviana* (L) SER. WHOLE PLANT

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ABSTRACT

Plants are of the best reservoirs of medicinal wealth among the natural medicines. The main aim of this study was to screen the phytochemicals and minerals present in the whole plant of *Mollugo cerviana* (L.) Ser. Preliminarily all the phytochemicals were extracted using different solvents (Aqueous, ethanol, methanol, acetone, chloroform, and petroleum ether) and screened qualitatively, which showed the presence of carbohydrates, proteins and amino acids, alkaloids, phenols, flavonoids, tannins, terpenoids, steroids, saponins, cardiac glycosides, gums and mucilages and thiols at higher concentration in the ethanolic extract than the other solvents tested. Depending on this, HPTLC, GC-MS, quantitative and mineral analysis were carried out in tha ethanolic extract of *Mollugo cerviana*. HPTLC analysis confirmed the presence of alkaloids, flavonoids and terpenoids, whereas GC-MS analysis revealed the presence of five bioactive compounds in the ethanolic extract. In addition to this quantitative estimation of this extract showed to contain alkaloids, total phenols, flavonoids, tannins and steroids and among this, total phenols were found to be present at higher quantity. Ethanolic extract of *Mollugo cerviana* was also found to possess minerals such as copper, zinc and magnesium at considerable amounts. Thus, ethanolic extract of *Mollugo cerviana* may be helpful for rationale use of this plant in the modern system of health care.

INTRODUCTION

Natural phytochemicals presented in herbs are natural bioactive compounds found in plants and these are divided into two groups; primary and secondary compounds have been widely used to cure, and reduce the risk of human diseases [1]**.** It has been reported that plant extracts are commonly used in traditional medicine and its contribution with respect to health coverage was estimated for over 80% of the world's population, especially in the developing world [2,3]. The bromatological and mineral analysis of edible plants plays an important role in assessing their nutritional significance.

The chosen medicinal plant *Mollugo cerviana* (L.) Ser. belongs to the family Molluginaceae, commonly known as threadstem carpet weed. *Mollugo cerviana* (L.)Ser. is a species of flowering plant known by the common name threadstem carpetweed. It can be found on most continents growing as a weed in many types of dry sandy habitat types. *Mollugo cerviana* (L.)Ser. known to cure diseases like blood impurity, burns, Gonorrhea, Hangover, jaundice, Pleurisy and effective in antifungal, antimicrobial, Diaphoretic, febrifuge, stomachic and laxative. The importance of medicinal plant in drug development is known to us and humans have used them for different diseases from the beginning of human history. Traditional folk treatment from wild plants has always guided researchers to search for novel medications to develop healthy life for humans and animals and also analysis of minerals from plants plays an important role in assessing their nutritional significance. In addition, some medicinal plants are still obscured within the plant which need to be scientifically evaluated [4,5]. The search for new pharmacologically active agents from natural resources such as plants, animals and microbes led to discovery of many clinically useful drugs. Hence, the present study was aimed to carry out phytochemical screening and mineral analysis of the whole plant extract of *Mollugo cerviana*.

MATERIALS AND METHODS

Collection of the plant Material

The whole plant of *Mollugo cerviana(L.)Ser.* was collected from Theni District, Tamil Nadu,India. The collected plant

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material was identified and authenticated at Botanical Survey of India, Tamilnadu Agricultural university, Coimbatore, Tamil Nadu, India. A voucher specimen of the plant was deposited at the herbarium of the BSI (Register Number: BSI/SRC/5/23/2016/Tech/1203).

Preparation of Extract

Whole plant material was collected freshly and washed in distilled water and allowed for shade dry and the dried sample was powdered. The powdered material (10 g) was extracted with 100ml of the selected solvents (Aqueous, ethanol, methanol, acetone, chloroform, and petroleum ether) using soxhlet apparatus and filtered. The filtrate was concentrated and dried under reduced pressure and controlled temperature

Qualitative Analysis

Qualitative phytochemical analysis of phytochemicals such as carbohydrates, proteins, alkaloids, flavonoids, steroids, terpenoids, tannins, saponins, cardiac glycosides, thiols, gum & mucilages were carried out in the aqueous, ethanol, methanol, acetone, chloroform and petroleum ether extracts of whole plant of *Mollugo cervania* (L.)Ser. using standard protocols [6,7].

HPTLC Fingerprint Profile

The HPTLC analysis of the ethanolic extract of *Mollugo cerviana* was carried out to confirm the presence of alkaloids, flavonoids and terpenoids. The plant extract was loaded in the pre-coated HPTLC plates (silica gel 60 F 254) CAMAG REPROSTAR 3 and plate size 10×10 cm. The TLC plates loaded with sample were kept in twin trough chamber which was saturated with solvent vapors. HPTLC was carried out with toluene: ethyl acetate: diethylamine (7:2:1), chloroform: methanol:formic acid (8.5:1.0:0.5), and butanol: isopropyl alcohol (1:1) as a mobile phase for alkaloids, flavonoids, and terpenoids, respectively. The developed plates were dried by hot air oven at 60°C for 5 min to make the solvent evaporate from the plate. After drying, the plates were taken for photo documentation (CAMAG TLC Scanner) chamber. The images were taken at UV 254nm and UV 366 nm.

GC-MS (Gas Chromatography-Mass Spectrometry) Analysis

The phytochemical investigation of ethanolic extract of *Mollugo cerviana* was performed on a GC-MS equipment (ThermoScientific Co.) Thermo GC-TRACE ultra ver.: 5.0, Thermo MS DSQ II. Experimental conditions of GC-MS system were as follows: DB 5 -MS Capillary Standard Non - Polar Column, dimension: 30Mts, ID: 0.25 mm, Film thickness: 0.25µm. Flow rate of mobile phase (carrier gas: He) was set at 1.0 ml/min. In the gas chromatography part, temperature programme (oven temperature) was 70°C raised to 260°C at 6°C/min and injection volume was 1 µl. Samples dissolved in ethanol were run fully at a range of 50-650 m/z and the results were compared by using Wiley Spectral library search programme.

Quantification of Phytochemicals

Quantification of alkaloids, total phenols, flavonoids, tannins, terpenoids and steroids were carried out.

Determination of Alkaloids

The plant extract (1mg) was dissolved in dimethyl sulphoxide (DMSO), added 1ml of 2N hydrochloric acid and filtered. This solution was transferred to a separating funnel, 5 ml of bromocresol green solution and 5 ml of phosphate buffer were added. The mixture was shaken with 1, 2, 3 and 4 ml chloroform by vigorous shaking and collected in a 10-ml volumetric flask and diluted to the volume with chloroform. A set of reference standard solutions of caffeine (20, 40, 60, 80 and 100 µg/ml) were prepared in the same manner as described earlier. The absorbance for test and standard solutions were determined against the reagent blank at 470 nm with an UV-Visible spectrophotometer. The total alkaloid content was expressed as mg of CE / g of extract [8,9].

Determination of total Phenolic Content

The concentration of phenolics in plant extract was determined using spectrophotometric method [10,11,12,13]. Folin-Ciocalteu assay method was used for the determination of the total phenol content. The reaction mixture consists of 1 ml of extract and 9 ml of distilled water was taken in a volumetric flask (25 ml). 1.0ml of Folin -Ciocalteu phenol reagent was treated to the mixture and shaken well. After 5 minutes, 10 ml of 7 % Sodium carbonate (Na_2CO_3) solution was treated to the mixture. The volume was made up to 25 ml. A set of standard solutions of gallic acid (20, 40, 40, 60, 80 and 100 µg/ml) were prepared in the same manner as described earlier. Incubated for 90 min at room temperature and the absorbance for test and standard solutions were determined against the reagent blank at 550 nm with an UV-Visible spectrophotometer. Total phenol content was expressed as mg of GAE/g of extract.

Determination of Flavonoid Content

Total content of flavonoid was measured by the aluminum chloride colorimetric assay[14,15,16,17].The reaction mixture consists of 1 ml of extract and 4 ml of distilled water was taken in a 10 ml volumetric flask. To the flask, 0.30 ml of 5 % sodium nitrite was treated and after 5 minutes, 0.3 ml of 10 % aluminum chloride was mixed. After 5 minutes, 2 ml of 1M Sodium hydroxide was treated and diluted to 10 ml with distilled water. A set of reference standard solutions of quercetin (20, 40, 60, 80 and 100 µg/ml) were prepared in the same manner as described earlier. The absorbance for test and standard solutions were determined against the reagent blank at 510 nm using UV-Visible spectrophotometer. The total flavonoid content was expressed as mg of QE/g of extract.

Determination of Tannin Content

The tannins were determined by Folin - Ciocalteu method [18,19,201,21]. About 0.1 ml of the sample extract was added to a volumetric flask (10 ml) containing 7.5 ml of distilled water and 0.5 ml of Folin-Ciocalteu phenol reagent, 1 ml of 35 % Na2CO3 solution and dilute to 10 ml with distilled water. The mixture was shaken well and kept at room temperature for 30 min. A set of reference standard solutions of gallic acid (20, 40, 60, 80 and 100 µg/ml) were prepared in the same manner as described earlier. Absorbance for test and standard solutions were measured against the blank at 725 nm with an UV-Visible spectrophotometer. The tannin content was expressed in terms of mg of GAE /g of extract.

Determination of Steroid Content

Steroids were determined by Ferric chloride colorimetric method[22]. To the centrifuge tube added 1.0ml of the plant extract with 4.9ml of $FeCl_3$ precipitating reagent and centrifuged. To 2.5ml of the supernatant, 2.5ml of $FeCl_3$ diluting reagent and 4.0ml of concentrated H_2SO_4 was added. A blank was also prepared simultaneously by taking 5.0ml of diluting reagent . Then add 4.0 ml of conc.H_2SO_4 to each tube. A set of standards (0.5-2.5ml) were taken and made upto 5.0ml with $FeCl_3$ diluting reagent. Then added 4ml of conc. H_2SO_4. After 30 minutes of time interval the formation of pink color was developed and the intensity was read at 540nm against a reagent blank. The amount of steroid in the sample is expressed as mg of cholesterol equivalent/g of plant extract.

Quantitative Determination of Selected Minerals

Quantitative determination of selected minerals like Copper, Zinc and Magnesium were analyzed by Atomic Absorption Spectrophotometer (AA 6300, Shimadzu, Japan).

Statistical Analysis

Statistical analysis was carried out in triplicates (n=3), and standard error of the mean (SEM) was calculated. All the data were analyzed using analysis of variance with the statistical software Prism 7.0 version.

RESULTS AND DISCUSSION

The analysis and characterization of bioactive compounds from plants is important to ascertain their medicinal value [23].

Qualitative Phytochemical Screening

The qualitative phytochemical analysis of various solvent extracts of *Mollugo cerviana* is given in Table 1.

Table 1 Qualitative phytochemical analysis of different solvent extracts *Mollugo cerviana* (L.) Ser

S. No.	Phytochemicals	Aqueous	Ethanol	Methanol	Acetone	Chloroform	Petroleum ether
1.	Carbohydrates	++	+++	+	+	+	+
2.	Proteins and amino acids	++	+++	++	++	-	+
3	Alkaloids	++	+++	+++	++	+	+
4	Flavonoids	++	+++	+++	++	++	++
5	Phenols	++	++	++	++	-	+
6	Steroids	++	++	+	+	+	-
7	Terpenoids	++	+++	+++	+	+	+
8	Tannins	++	++	++	+	+	+
9	Saponins	++	++	+	+	-	-
10	Cardiac glycosides	+	++	+	+	-	+
11	Gum & Mucilages	+	+	+	-	-	-
12	Thiol's	+	+	+	-	+	-

+ → present in small concentration; ++ → present in moderately high concentration; +++ → present in very high concentration; - → absent.

It was observed that aqueous, ethanol and methanol extracts revealed the presence of all the phytochemicals that were tested, such as carbohydrates, proteins, and amino acids, alkaloids, phenols, flavonoids, tannins, terpenoids, steroids, saponins, cardiac glycosides, gum and mucilages and thiols. While, Acetone, chloroform and petroleum ether extracts revealed the occurrence of most of the phytochemicals tested, at the mean while, it has also been exhibited absence of fewer phytochemicals that were tested (Table 1). Among all the solvents were used for the extraction and phytochemical analysis of *Mollugo cerviana* plant, ethanolic extract were showed to contain all the phytochemicals at higher concentrations. Ethanol was shown to be the most excellent solvent, as polar solvents have greater capability to extract the maximum phytoconstituents than other nonpolar solvents. Hence, ethanol was showed to be penetrating the cellular membrane rapidly to extract the intracellular components from the plant origin[24].

Roghini and Vijayalakshmi[25] reported that *Citrus paradisi* plant extract in various solvents were showed the presence of phytochemicals such as alkaloids, carbohydrates, saponins, reducing sugars, flavonoids, phenols, proteins, tannins, terpenoids and glycosides. Particularly these phytochemicals were found to be higher concentration in ethanolic extract than the other solvent extracts tested.

Carbohydrates are biological macromolecules serve as a source of energy and also possess antioxidant activity against reactive oxygen species, chronic and degenerative diseases. Glycosylated natural products have been commonly utilized as antimicrobial drugs and now as emerging anti-cancer drug candidates[26].Proteins are reported to possess antioxidant activity [27]and amino acids are involved in the synthesis of proteins, amines, alkaloids, vitamins, enzymes and terpenoids[28]

Alkaloids have many pharmacological activities including antihypertensive effects, antiarrhythmic effect, antimalarial activity, and anticancer actions[29] . Phenolic compounds could be served as free radical scavengers and protect oxidative damages. Anti-ulcer, anti-inflammatory, antioxidant, cytotoxic, antitumor, antispasmodic, antidepressant activities and antidiabetic effects are another biological activities of phenolic compounds. They are also responsible for the protection of tissue membranes and proteins against harmful free radicals[30,31,32]. Flavonoids stated to possess many useful properties, including anti-inflammatory activity, enzyme inhibition, antimicrobial activity, oestrogenic activity, anti-allergic activity, antioxidant activity, vascular activity, cytotoxic and antitumor activity[33,34] Tannins have an amazing stringent properties. They are known to hasten the healing of wounds and inflamed mucous membranes. Recently the tannins have attracted scientific interest, especially due to the increased incidence of deadly illnesses such as AIDS and various cancers[35,36]. Terpenoids play an important role in traditional herbal remedies and are under investigation for antibacterial, anti neoplastics, and other pharmaceutical functions. There are many different classes of naturally occurring compounds[37,38,39]. Steroids have been reported to have antibacterial properties [40] and they are very important compounds especially due to their relationship with compounds such as sex hormones [41]. Saponins used in hypercholesterolemia, hyperglycemia, antioxidant, anti-cancer, anti-inflammatory, anti-fungal central nervous system activities and weight loss. Saponins reduce the uptake of certain nutrients including glucose and cholesterol at the gut through intra lumenal physicochemical interaction. Hence, it has been reported to have hypocholesterolemic effects and thus, they may aid in lessening the metabolic burden that would have been placed on the liver[42,43]. Gums and mucilages of different sources and their derivatives represent a group of polymers widely used in pharmaceutical dosage forms and are useful as tablet binders, disintegrants, emulsifiers, suspending agents,

gelling agents, stabilizing agents, thickening agents, film forming agents in transdermal and periodontal films, buccal tablets as well as sustaining agents in matrix tablets and coating agents in microcapsules including those used for protein delivery[44].

HPTLC analysis

HPTLC analysis of alkaloids in ethanolic plant extract was presented in **Plate 1, Figure 1 and Table 2.**

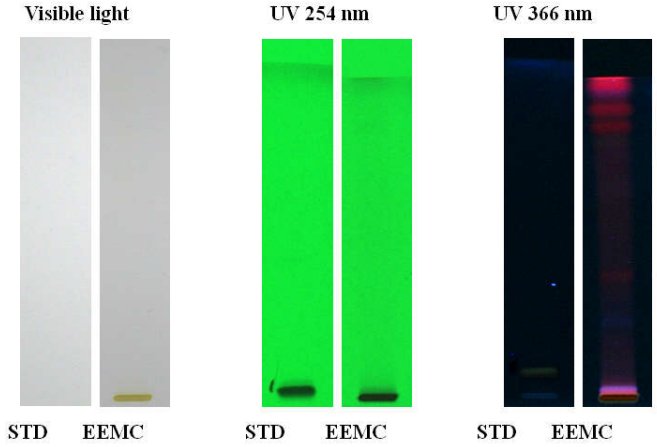


Plate 1 HPTLC chromatogram of alkaloids of ethanolic extract of *Mollugo cerviana*

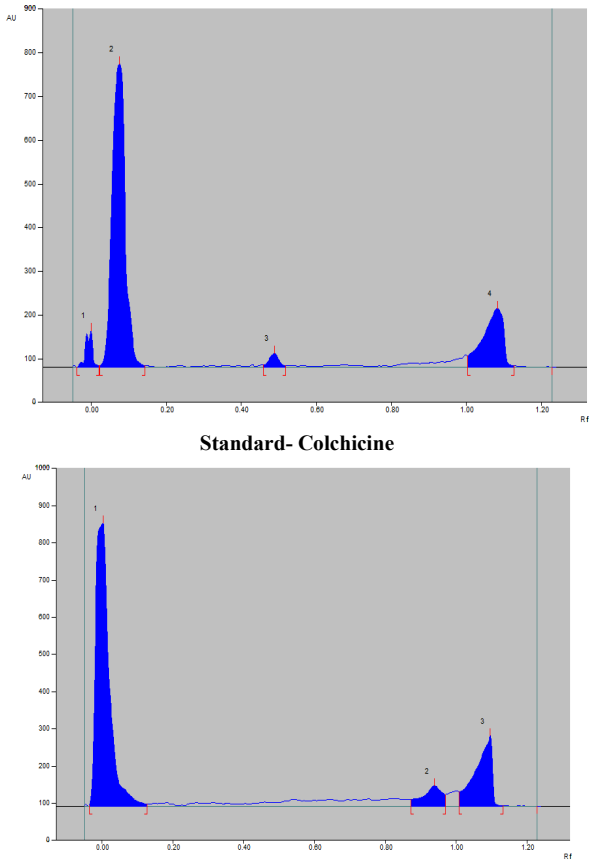


Figure 1 Peak densitogram display of alkaloids of ethanolic extract of *Mollugo cerviana* (Scanned at 254nm)

Table 2 HPTLC – peak table of alkaloids of ethanolic extract of *Mollugo cerviana*

Track	Peak	Rf value	Height	Area	Assigned substance
Standard	1	0.14	3.5	17438.6	Colchicine
Sample	1	0.13	6.1	21276.2	Alkaloid 1
Sample	2	0.97	31.9	2140.0	Unknown
Sample	3	1.13	1.3	6229.4	Alkaloid 2

The Rf value of the different compounds present in the ethanolic plant extract was showed to be 0.13, 0.97 and 1.13 of peak 1, 2 and 3, while, the Rf value of standard Colchicine was showed to be 0.14 at peak 1. These observed regions confirmed the presence of two alkaloids compounds in the plant extract.

In accordance with our results Chavan *et al*[45], found that methanolic extract of *Cassia fistula* L. leaves confirmed that alkaloids are the chief group of phytochemical in the plant extract. HPTLC analysis of flavonoids in the ethanolic plant extract was presented in Plate 2, Figure 2 and Table 3.

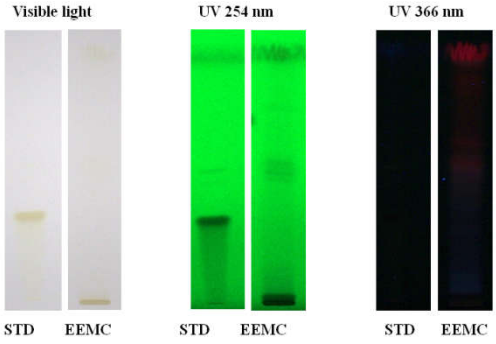


Plate 2 HPTLC chromatogram of flavonoids of ethanolic extract of *Mollugo cerviana*

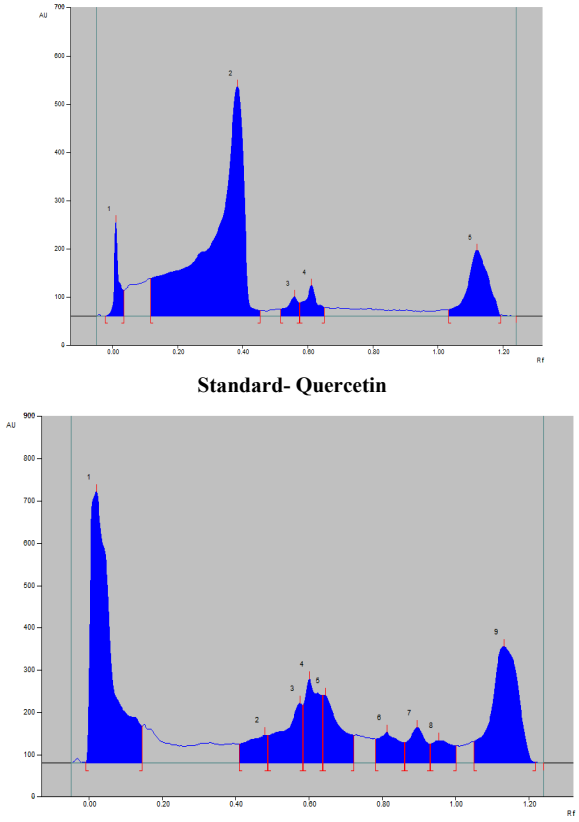


Figure 2 Peak densitogram display of flavonoids of ethanolic extract of *Mollugo cerviana* (Scanned at 254 nm)

Table 3 HPTLC – peak table of flavonoids of ethanolic extract of *Mollugo cerviana*

Track	Peak	Rf value	Height	Area	Assigned substance
Standard	1	0.45	10.9	31681.7	Quercetin
Sample	1	0.15	87.0	25920.1	Flavonoid 1
Sample	2	0.49	64.9	2723.9	Flavonoid 2
Sample	3	0.58	135.9	5451.7	Unknown
Sample	4	0.64	159.1	5564.4	Unknown
Sample	5	0.72	65.6	5428.1	Unknown
Sample	6	0.86	47.8	2949.1	Unknown
Sample	7	0.93	44.6	2710.7	Unknown
Sample	8	1.00	40.7	2096.9	Unknown
Sample	9	1.22	1.4	14365.6	Flavonoid 3

The Rf value of the varying compounds exist in the plant extract was found to be 0.15, 0.49, 0.58, 0.64, 0.72, 0.86, 0.93, 1.00 and 1.22 of peak 1,2,3,4,5,6,7,8 and 9, whereas, the Rf value of standard Quercetin was found to be 0.45 at peak1. These obtained regions confirmed the presence of three flavonoid compounds in the ethanolic plant extract.

Similar to our results Rajamani *et al*[46] have found that ethanolic extract of Orthodox black tea possessed six different types of flavonoid compounds.

HPTLC analysis of terpenoids in the ethanolic extract of *Mollugo cerviana* is depicted in Plate 3, Figure 3 and Table 4.

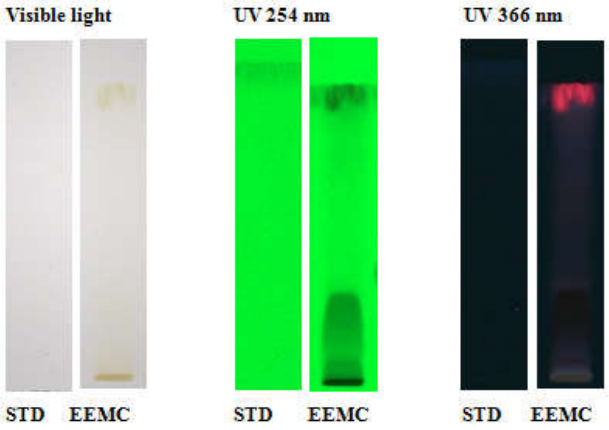
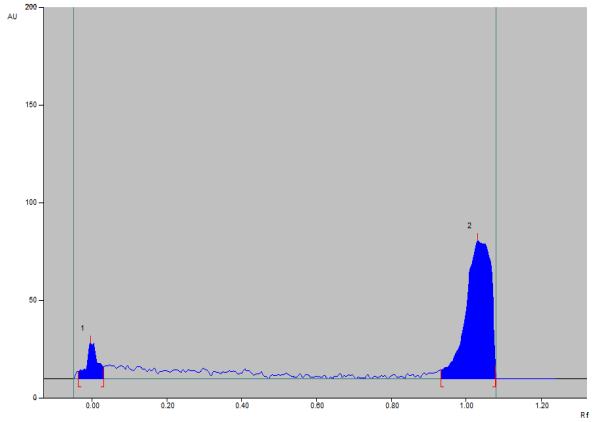
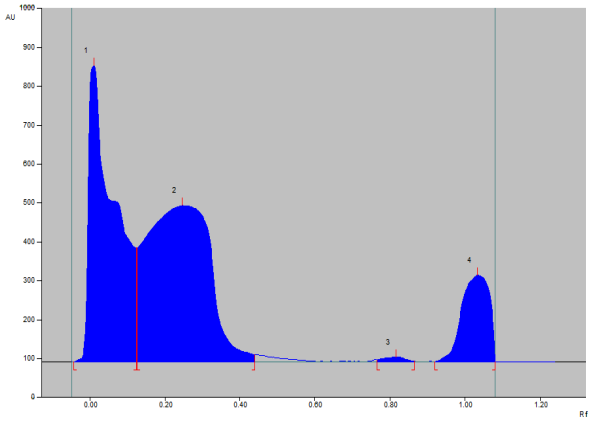


Plate 3 HPTLC chromatogram of terpenoids of ethanolic extract of *Mollugo cerviana*



Standard - Lupeol



Mollugo cerviana plant extract

Figure 3 Peak densitogram display of terpenoids of ethanolic extract of *Mollugo cerviana* (Scanned at 254nm)

Table 4 HPTLC – peak table of terpenoids of ethanolic extract of *Mollugo cerviana*

Track	Peak	Rf value	Height	Area	Assigned substance
Standard	1	1.08	6.6	3368.1	Lupeol
Sample	1	0.12	293.1	38088.6	Unknown
Sample	2	0.44	18.5	50175.5	Unknown
Sample	3	0.87	1.4	538.1	Terpenoid 1
Sample	4	1.08	0.0	11937.5	Lupeol

The Rf value of varying compounds present in the ethanolic plant extract was found to be 0.12, 0.44, 0.87 and 1.08 of peak 1, 2, 3 and 4, whereas, the Rf value of standard Colchicine was found to be 1.08 at peak1. These regions revealed the presence of one Lupeol and one terpenoid compound in the plant extract. Similar findings were given by Malviya and Dwivedi [47], who reported that *Ailanthus excels* extract showed to possess terpenoid compound. Chanu *et al* [48], also reported that *Parkia javanica* HPTLC profile of plant extract showed the presence of terpenoids.

GC-MS Analysis

Analysis of bioactive compounds using GC-MS technique is the leading one to determine long chain hydrocarbons, alcohols, acids, esters, alkaloids, steroids, amino and nitro compounds, etc., [49]. Figure 4, Table 5 and Figure 5 showed the presence active compounds present in the ethanolic plant extract

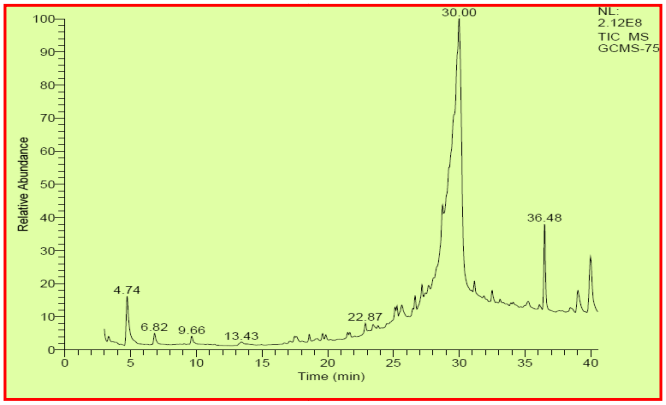


Figure 4 GC-MS Spectrum of ethanolic extract of *Mollugo cerviana* (L.) Ser.

Table 5 GC-MS analysis of the ethanolic extract of *Mollugo cerviana* (L.) Ser

S.No	Compound Name	% of Peak Area	Retension time (RT)	Molecular formula (MF)	Molecular weight (MW)
1.	Peroxide, dimethyl	6.11	4.74	$C_2H_6O_2$	62
2.	Cyclododecane	1.28	6.82	$C_{12}H_{24}$	168
3.	7-Hexadecene, (Z)	0.91	9.66	$C_{16}H_{32}$	224
4.	Hexadecanoic acid, ethyl ester	1.07	22.87	$C_{18}H_{36}O_2$	284
5.	Squalene	7.47	36.48	$C_{30}H_{50}$	410

GC – MS analysis of the plant extract exhibited the presence of five major peaks and the corresponding components to the peaks were determined as follows: Peroxide, dimethyl (6.11%), Cyclododecane (1.28%), 7-Hexadecene, (Z) (0.91%), Hexadecanoic acid, ethyl ester (1.07%) and 2,6,10,14,18,22-Tetracosahexaene,2,6,10,15,19,23-hexamethyl- (CAS) (7.47%) were present.

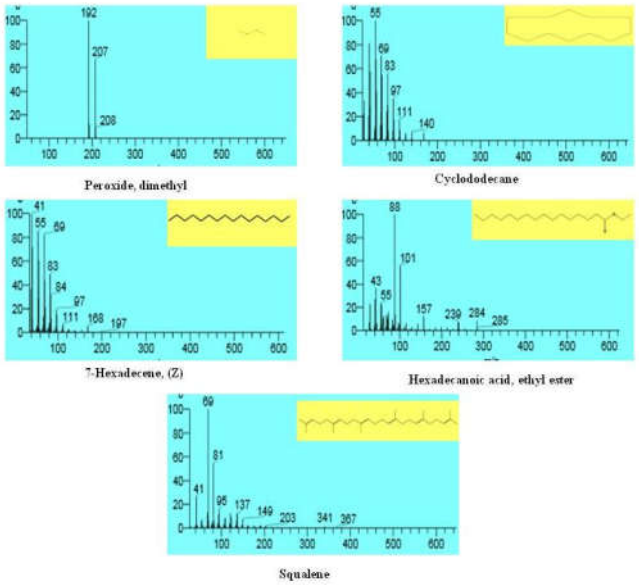


Figure 5 Mass spectra of identified compound from ethanolic extract of *Mollugo cerviana* (L.) Ser.

Dimethyl peroxide, generally known as peroxide dimethyl, originates from the class of organic compounds known as dialkyl peroxides. Dimethyl peroxide is a water soluble compound and highly weak basic (essentially neutral) compound (depends on its pKa) and is mainly located in the cytoplasm. Dimethyl peroxide may be present in green vegetables. Thus, it makes dimethyl peroxide as a potential biomarker for the consumption of this food product[50]. Cyclododecane was found to possess antimicrobial, antioxidant and antitumor activities[51]. Previous studies reported that 7-Hexadecene, (Z) found to have anti-bacterial activity[52]. Hexadecanoic acid, ethyl ester possessed antioxidant, flavor, hypocholesterolemic, nematicide, pesticide, lubricant, anti-androgenic, hemolytic activities and also act as 5-Alpha reductase inhibitor[53].

Squalene (triterpine) is a form of phenolic compound and found to be present in the latex and resins of many plants. Squalene found to possess activities like anti-microbial, chemopreventive against colon carcinogenesis[54]. Squalene also have been reported to exert antioxidant, anticancer, chemopreventive, hepatoprotective and gastro preventive effects, pesticide, and sunscreen properties and is also serve as a cosmetics in natural moisturizer[55]. Similar to our results, Velmurugan and Anand [56] carried out GC-MS analysis of ethanolic leaf extract of *Phyllodium pulchellum* and the results showed the presence of ten bioactive compounds. GC-MS analysis of the ethanolic extract of the whole plant *Drosera indica* showed to possess 8 active compounds [57].

Quantification of Phytochemicals

The importance of biological, chemical and pharmacological evaluation of plant-derived bioactive compounds used to cure numerous human ailments has been increasingly recognized in the last few decades, but still there are innumerable potentially useful medicinal plants and herbs waiting to be evaluated and exploited for their effective therapeutic application[58]. The results of quantitative determination of secondary metabolites in the ethanolic plant extract is given in Table 6

Table 6 Quantitative estimation of secondary metabolites of ethanolic extract of *Mollugo cerviana*

S.No.	Phytochemicals	QUANTITY (mg/g of crude plant extract)
1.	Alkaloids	42.33 ± 0.57
3.	Total phenols	95.66 ± 0.57
2.	Flavonoids	45.33 ± 1.52
4.	Tannins	46.66 ± 1.15
5.	Steroids	75.33 ± 1.15

Values are expressed in Mean ± SEM (n=3)

The secondary metabolites such as alkaloids (42.33 ± 0.57mg/g of Caffeine equivalent), total phenols (95.66 ± 0.57 mg/g of Gallic acid equivalent), flavonoids (45.33 ± 1.52mg/g of Quercetin equivalent), tannins (46.66 ± 1.15 mg/g of Gallic acid equivalent) and steroids (75.33 ± 1.15 mg/g of Cholesterol equivalent) were present in considerable quantities in the ethanolic extract of *Mollugo cerviana*. The plant extract was showed to possess total phenols at higher quantity followed by steroids.

Similarly, Tambe and Bhambar[59] estimated secondary metabolites in the *Hibiscus Tiliaceus* Linn. Wood Extracts and it was showed the presence of total phenolics (30.18±0.321mg/g), Tannins (83.03±0.029 mg/g), alkaloid (66.01±0.049 mg/g) and Flavonoids (91.01±0.046 mg/g). Likewise, *Hemidesmus indicus*, showed to contain total polyphenolic content at 45.5mg/g, total flavonoid at 27.3 mg/g respectively[60].

Quantitative Determination of Selected Minerals

Minerals and trace elements in medicinal plants play an important role due to its importance in several metabolic processes and prevent living organisms from several diseases[61]. The minerals such as zinc and magnesium are grouped as components of high priority for dietary consumption[62]. Minerals present in the ethanolic plant extract are given in Table 7.

Table 7 Quantity of minerals in the ethanolic extract of *Mollugo cerviana*

Element	Concentration(ppm)
Copper	0.16 ± 0.185
Zinc	0.05 ± 0.106
Magnesium	5.93 ± 0.053

Values are expressed in Mean ± SEM (n=3)

The results showed the presence of characteristic level of trace elements such as copper, zinc and magnesium in the plant extract. The quantified minerals in the plant extract were found to be present within permissible limits (FAO / WHO).

Our results are in accordance with the findings of Imo *et al*[63] who reported the presence of minerals such as, copper (0.075), zinc (0.422) and magnesium (5.297 ppm) in the ethanolic extract of *Xylopia aethiopica* fruits. Similarly, Dattaa *et al*[64] estimated minerals in the extract of *Amaranthus viridis* and the results showed the presence of minerals such as Cu (1.860 µg/g), Zn (0.019 mg/g) and Mg (0.039 mg/g) at considerable levels.

Copper is crucial to all living organisms and it is a key constituent of the respiratory enzyme complex. Symptoms of deficiency can produce anemia, bone abnormalities, impaired growth, abnormalities in glucose and cholesterol metabolism. It is very important for proper nerve function, bone growth, enhanced body to use of sugar and protection of cell membranes from destruction by free radicals[65]. **Zinc** plays an important role in more than 300 enzymes involved in synthesis and degradation of biomolecules, metabolism of other micronutrients[66]. It plays a role in immune function, protein synthesis, wound healing, DNA synthesis and cell division. It also supports normal growth and development during pregnancy, childhood, and adolescence [67] Magnesium dilates and relaxes the smooth muscles of the bronchi. Magnesium deficiency can increase the risk of osteoporosis, the risk of atherosclerosis and lead to oxidative stress[68,69].

The role of minerals in fighting cancer is a constantly-evolving field of research, with promising data that the nutrients we eat can help fight the development or progression of cancer. Minerals are needed by the cancer patient in order to prevent and to increase the chances for natural immunological remission of cancer. If these minerals are taken in proper amounts these can be helpful in cancer therapy. In addition, taking mineral supplements may decrease the efficacy of some pharmaceuticals [70].

CONCLUSION

In the present study, phytochemical screening of different solvent extracts of (aqueous, ethanol, methanol, acetone, chloroform and petroleum ether) *Mollugo cerviana* was showed to possess various phytochemicals such as carbohydrates, proteins, alkaloids, phenolic compounds, flavanoids, tannins, terpenoids, steroids, saponins, cardiac glycosides, thiols, gum & mucilages. Among the solvents tested, ethanol was found to be a better solvent to solubilize all the phytochemicals that were tested. Hence, ethanolic plant extract was selected for advance studies. HPTLC analysis of the plant extract confirmed the occurrence of alkaloids, flavonoids and terpenoids. GC – MS analysis showed the presence of five major active components with an effective medicinal value. Quantitative estimation of plant extract was showed to possess considerable quantity of secondary metabolites such as, alkaloids, total phenols, flavonoids, tannins and steroids and also this plant found to contain minerals like copper, zinc and magnesium in the considerable levels. Thus, the ethanolic extract of *Mollugo cerviana* whole plant may serve as a promising therapeutic agent and can be beneficially used for their medicinal properties to treat various emerging diseases.

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