Research Article

Phytochemical Evaluation and Metabolic Profiling of Methanolic Extracts from the Stem and Leaf of *Suregada multiflora* (A. Juss.) Baill.: An Ethnobotanically Important Medicinal Plant

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ABSTRACT

Aim: In Indian traditional knowledge; many plants are documented for their therapeutic usage. Secondary metabolites are responsible for this pharmacological potentiality. *Suregada multiflora*, known as false lime, is used traditionally for the treatment of many diseases. Therefore, the present study was undertaken to evaluate the phytochemical profile of stem and leaf methanolic extracts from *S. multiflora* to substantiate their therapeutic efficacy. **Materials and Methods:** Physicochemical evaluation was undertaken to check its usage in crude drug formulations; it was followed by the phytochemical analysis of the stem and leaf using non-polar to polar solvents. Methanolic extracts of stem and leaf were subjected to HPTLC analysis, and to identify the compounds, LCMS analysis was carried out. **Results:** Preliminary phytochemical analysis showed that the methanolic extract of stem and leaf comprises a remarkable number of phytoconstituents. It was further substantiated with HPTLC analysis in terms of the total number of peaks, peak heights, peak area, percent area and R*f* values. Following this, LC-MS analysis was conducted, and positive ionization of the leaf revealed 47 components, while the negative mode showed 60 identified molecules. Similarly, the positive ionization of the stem showed 41 components, and the negative ionization of the stem showed 58 identified components. **Conclusion:** Methanolic extract of *S. multiflora* stem and leaf contains a pool of phytochemicals which might be accountable for its therapeutic value and justifies its traditional usage.

Keywords: *Suregada multiflora*, Qualitative Phytochemistry, Physico-chemical standardization, Spectrophotometry.

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INTRODUCTION

Generally, the focus on using medicinal herbs had been on disease treatment rather than prevention. However, there have been reports from the literature that the use of medicinal plants and the substances found in them in preventing diseases. Traditional Knowledge (TK) has long been used in primary healthcare, but it must be thoroughly examined for therapeutic applications and possible toxicity. Traditional practitioners have used medicinal plants as antibiotics. Additionally, numerous types of research revealed gaps in therapeutic efficacy against illnesses. Analytical examinations of herbals are experiencing vast expansion in the last several years. Currently, phytochemicals profiling by LC-MS/GC-MS emerged as a powerful analytical technique. Using HR-LCMS, bioactive compounds from many plant species like *Pongamia pinnata, Gardenia resinifera, Dodonea viscosa* and *Gymnospora emarginata* were analyzed and reported. Single liquid chromatography–mass-spectrometric analysis of samples records thousands of data entries. Rigveda refers to three types of species such as Vriksha, Osadhi and Virudh. In Atharvaveda shape and morphology of herbals are described,

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while Yajurveda records four groups of medicinal species.[1] A proactive approach to healthcare that places emphasis on prevention at various points along with the healthcare continuum i.e., health promotion, disease prevention, and chronic illness management. This is done using medicinal plants, and one such plant selected for the present work is *Suregada multiflora*. Members of Euphorbiaceae are extensively distributed in tropical and subtropical regions of Asia and Africa. In Thai traditional medicine, bark of *Suregada multiflora* were used to cure hepatitis, skin conditions, lymphatic disorders, fungal infections and leprosy. The wood was used to treat pyrexia, dermatitis, and venereal conditions. The roots were also used to treat skin infections and lymphatic disorders.[2] According to reports, this plant contains anti-HIV properties against the herpes simplex virus. Therefore, there is ample scope in this Indian medicinal plant to unravel many human ailments/ disorders. Hence, the present study has been done with the objective of evaluating the secondary metabolites in *Suregada multiflora* using spectrophotometric techniques to substantiate its application in traditional usage.

MATERIALS AND METHODS

Collection and processing of plant material

The plant material *S. multiflora* was collected from Thiruvananthapuram, and its taxonomic identity was confirmed by the Curator, Department of Botany, University of Kerala. The stem and leaf of the plant were separately washed, shade-dried, and ground to obtain a coarse powder. Finally, the plant parts were separately subjected to Soxhlet extraction using different solvent systems based on their polarity.

Physico-chemical and preliminary phytochemical analysis

Physiochemical parameters were performed as per the standard method of quality control methods for medicinal plant materials. Phytochemical screening was carried out on the crude extracts using standard procedures.[3]

High-Performance Thin Layer Chromatography (HPTLC) analysis

Using an Automatic TLC Sampler 4 (ATS4) and a CAMAG microliter syringe, spots of the plant extracts were placed on the plate. About 5 µL and 10 µL of the extracts were applied on the TLC plate and developed in the toluene:ethyl acetate solvent system (5:1.3). The formed plate was dried by air and observed at UV wavelengths of 254, 366, and 575 nm.

Liquid Chromatography-Mass Spectrometry (LC-MS) studies

A binary pump-equipped Mariner Bio spectrometer was used to conduct the LC-MS analysis. A Q-TOF mass spectrometer with an ESI source was connected to the HPLC. To conduct the analysis, 5µL of the extracts were put into the analytical column. Utilizing a spectrum database for organic molecules in the SDBS programme, the mass fragmentations were identified.

RESULTS

Physico-chemical and preliminary phytochemical analysis in *Suregada multiflora*

The physicochemical analysis of the stem and leaf powder of *S. multiflora* was carried out and depicted in Table 1. Loss on Drying (LOD) at 105°C for stem and leaves were 7.619 and 10.509, respectively, indicating less chance of bacterial growth.

Phytochemical investigation of stem and leaf extracts (hexane; chloroform; ethyl acetate; methanol, and water) of *S. multiflora* revealed the presence of major secondary metabolites like carbohydrates, proteins, alkaloids and terpenoids. Quinones, coumarins, and flavonoids were totally absent in the extracts. The results of the same were depicted in Table 2.

HPTLC fingerprinting of stem and leaf methanolic extract of *S. multiflora*

Since most of the phytoconstituents were resolved in methanol, methanolic extracts of stem and leaf were subjected to HPTLC analysis. The chromatograms were obtained upon scanning at UV 254 and 366 nm, and under white light after derivatization (Figure 1). The peak height, peak area, percent area and R_f values

Table 1: Physico-chemical evaluation of *S. multiflora* in stem and leaf powder.

Parameters	Stem	Leaf
LOD at 105°C	7.619	10.509
Total Ash (% w/w)	2.56	5.555
Acid insoluble ash	0.1	0.285
Water soluble ash	1.4	2.805
Sulphated ash	4.58	9.0057
pH	4.85	4.38
Volatile oil	Nil	traces
Alcohol soluble extractives	6.077	23.63
Water soluble extractives	8.178	21.03
Swelling index (mL)	4.5	6
Foaming index	200	142.85

Table 2: Preliminary phytochemical analysis of different solvent extracts of *S. multiflora* stem and leaf.

Tests	Stem extracts					Leaf extracts				
	1	2	3	4	5	1	2	3	4	5
Saponins	-	-	-	-	+	-	-	-	+	+
Tannins	-	-	-	-	+	-	-	-	-	+
Terpenoids	+	+	+	+	-	+	+	-	-	-
Phenols	-	-	+	+	-	-	-	+	++	-
Steroids	-	-	-	-	-	+	+	+	-	-
Glycosides	-	-	+	+	-	-	+	+	+	+
Carbohydrates	-	-	-	+	+	+	+	+	+	+
Alkaloids	+	+	+	+	+	+	+	+	+	+
Flavanoids	-	-	-	-	-	-	-	-	+	-
Coumarins	-	-	-	-	-	-	-	-	-	-
Proteins	-	-	+	+	+	-	-	-	+	+
Quinones	-	-	-	-	-	-	-	-	-	-

(Hexane; 2. Chloroform; 3. Ethyl acetate; 4. Methanol; 5. Water + positive and – negative)

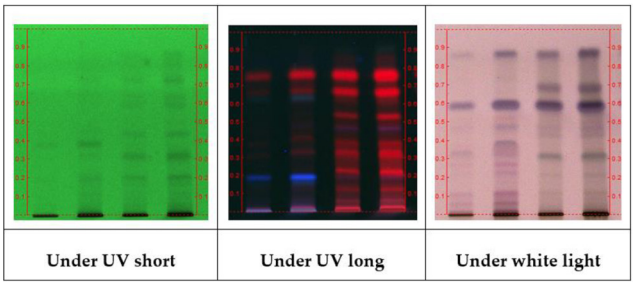


Figure 1: HPTLC chromatogram of methanolic extracts of stem and leaf of *S. multiflora.*

of the unknown substances were depicted. When the densitogram is scanned at 254 nm, it shows 3 and 9 peaks for 5 and 10 µL of stem extract and 10 and 12 peaks for leaf extract (Figure 2). In contrast, when the densitogram is scanned at 366 nm, it shows 4 and 8 peaks for 5 and 10 µL, respectively, for the stem extract and 11 and 10 peaks for the leaf extract (Figure 3). Similarly, the densitogram scanned at 575 nm represents 8 and 12 peaks for 5 and 10 µL respectively for stem extract, and 8 and 9 peaks with 5 and 10 µL, respectively for leaf extract (Figure 4). The R_f values and area% for the phytoconstituents found in the tested samples were also computed.

LCMS analysis of stem and leaf methanolic extracts of *S. multiflora*

The LC-MS chromatogram of stem and leaf methanolic extract of *S. multiflora* was conducted and shown in Figures 5 and 6. The positive ionization of stem revealed 41 compounds, of which 18 showed abundance (Table 3), and the negative ionization of stem showed 58 compounds with 37 showed abundance.

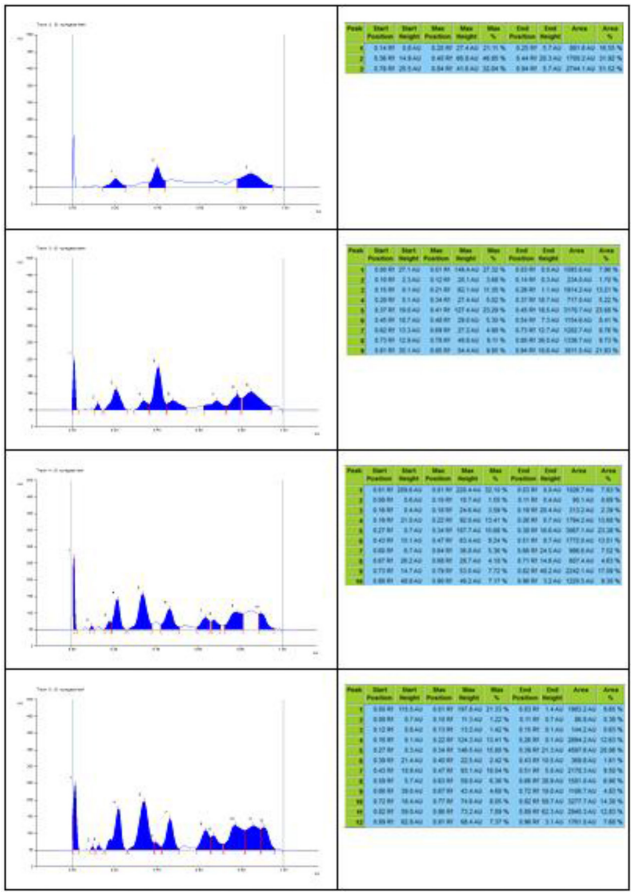


Figure 2: HPTLC fingerprints representing R_f value and area % of stem and leaf extracts at 254 nm.

The positive ionization of leaf methanolic extract revealed 47 compounds and 19 were abundant (Table 4). The negative ionization of leaf methanolic extract revealed 60 compounds of which 28 were abundant.

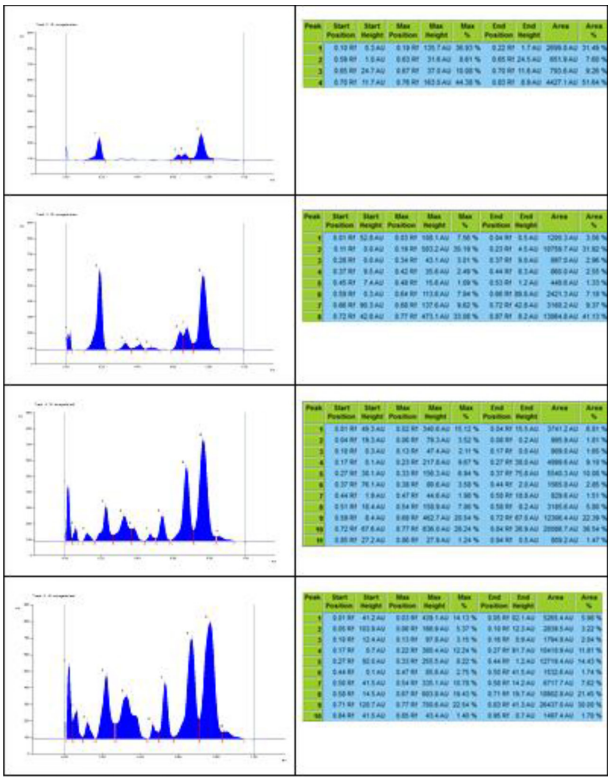


Figure 3: HPTLC fingerprints representing R_f value and area % of stem and leaf extracts at 366 nm.

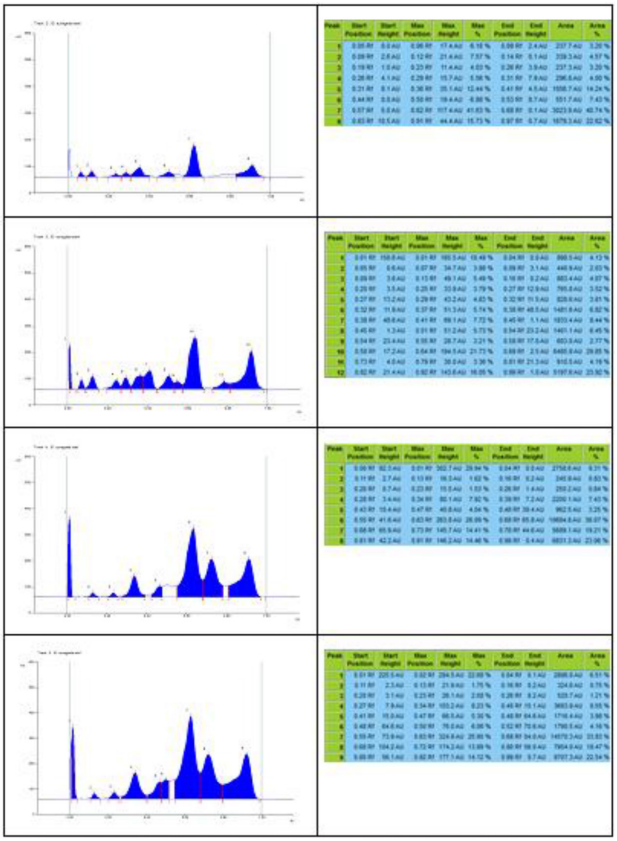


Figure 4: HPTLC fingerprints representing R_f value and area % of stem and leaf extracts at 575 nm.

Interestingly, compounds such as N-ethylglycocyamine, terbinafine, actinidine, cinncassiol C3, euphornin were common in stem and leaf-positive mode, while benzoic acid, hamamelose, nicotiflorin, (-)-pinoresinol glucoside, corchorifatty acid F, loteprednol, 9,10-dihydroxy-12,13-epoxyoctadecanoate, 9-HOTE, 8S-HODE, phorbol 13 acetate 12-myristate were common in stem and leaf negative mode. No common compounds were noticed among stem-negative and stem-positive or leaf-negative and leaf-positive data.

DISCUSSION

Traditional medicine has employed the herb *Suregada multiflora* to cure a number of diseases. Owing to the multifarious potentialities of the plant, many researchers are encouraged to conduct scientific studies on this ethnobotanically important medicinal plant. The illicit addition of pharmaceutical compounds or their analogues and the misidentification of crude pharmaceuticals might result in major health issues, and hence a robust regulatory oversight is the need of the hour to protect the public. Pharmacological studies are the most reliable, accurate and inexpensive means to evaluate plant drugs. The purpose of the current investigation is to elucidate the main components of *S. multiflora* and identify those that have pharmacological activity. Analysis of physicochemical parameters predicts the probability of plants being used in the preparation of Ayurvedic formulations. The loss on drying value of both the stem and leaf powder of *S. multiflora* falls within the recommended range of 8%-14% for vegetable drugs, which implies that the plant can be stored for a long period of time with less probability of microbial attack. Evaluation of physico-chemical characteristics is essential for determining the presence of adulterants and improper handling of drugs. To pinpoint the identity and purity of crude drugs, especially in powder form ash values can be considered as quantitative standards. Moreover, the total ash content of crude drug reveals the care taken in drug preservation, the purity of crude and the prepared drug. Ash insoluble in acid is a part of total ash and measures the amount of silica present, especially the sand and siliceous earth. It is helpful for evaluating crude pharmaceuticals because it provides insight into the types of chemical ingredients that are present in them and is helpful for estimating chemical constituents that are soluble in the extraction solvent. Further, the volatile oil was found in traces in leaf powder and absent in the stem. Extractive values were high in leaf powder, indicating that the leaf could dissolve more constituents than the stem powder,

Table 3: List of abundant compounds in stem methanolic extract of *S. multiflora* in positive mode.

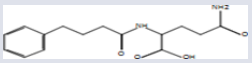
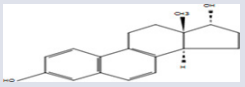
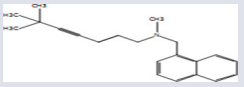
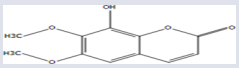
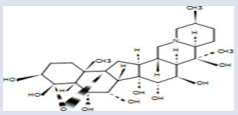
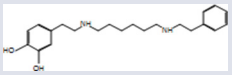
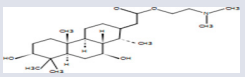
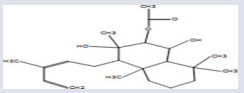
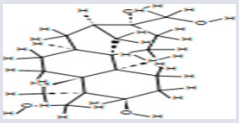
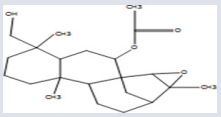
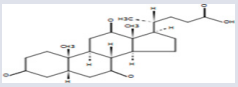
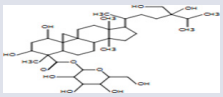
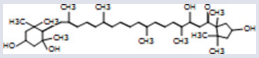
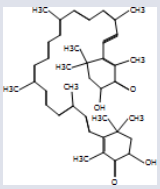
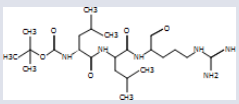
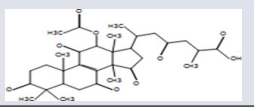
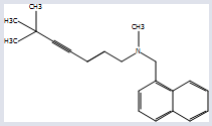
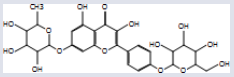
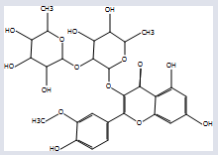
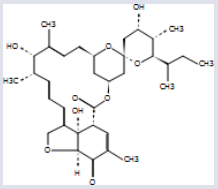
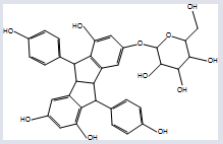
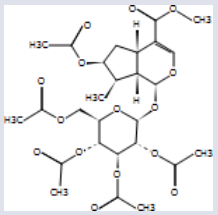
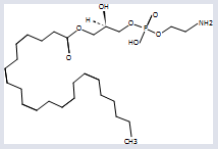
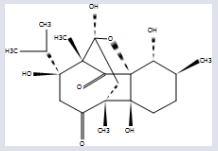
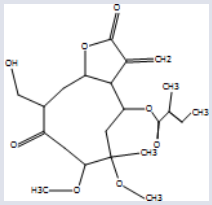
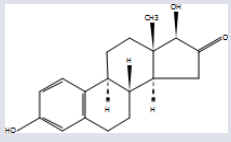
Name	Formula	Score	Mass	RT	Abundance	Compound structure (only Abundance)
(2S)-2-{[1-(R)-Carboxyethyl]amino} pentanoate	$C_8 H_{15} N O_4$	76.46	189.0989	1.727	328666	
Phenyl butyryl glutamine	$C_{15} H_{20} N_2 O_4$	32.48	292.1378	3.348	109208	
17alpha-Dihydroequilenin	$C_{18} H_{20} O_2$	54.81	268.1436	3.721	116925	
Terbinafine	$C_{21} H_{25} N$	59.76	291.203	3.773	136992	
Fraxidin	$C_{11} H_{10} O_5$	95.33	222.052	5.79	126357	
Protoverine	$C_{27} H_{43} N O_9$	76.61	525.2938	7.436	89954	
Dopexamine	$C_{22} H_{32} N_2 O_2$	82.62	356.247	8.041	592974	
Cassaidine	$C_{24} H_{41} N O_4$	83.29	407.3009	8.392	264130	
7-O-Acetylaustroinulin	$C_{22} H_{36} O_4$	65.38	364.2571	10.06	588812	
2R-hydroxy-stearic acid	$C_{18} H_{36} O_3$	96.05	300.2673	10.6	554099	
Aphidicolin	$C_{20} H_{34} O_4$	75.03	338.2439	11.31	1195865	
Epoxysiderol	$C_{22} H_{34} O_4$	62.84	362.2411	12.59	232670	
3,7,12-Trioxo-5β-cholan-24-oic Acid	$C_{24} H_{34} O_5$	59.05	402.2363	12.99	597648	
Cyclopassifloside II	$C_{37} H_{62} O_{11}$	69.41	682.4237	14.89	199519	
(3S,3'S,5R,5'R,6R)-6,7-Didehydro-5,6-dihydro-3,3',5,8'-tetrahydroxy-beta, kappa-caroten-6'-one	$C_{40} H_{56} O_5$	79.71	616.4153	14.89	188992	
7,8-Dehydroastaxanthianthin	$C_{40} H_{50} O_4$	88.01	594.3715	14.95	213530	
N-tert-Butyloxycarbonyl-deacetyl-leupeptin	$C_{23} H_{44} N_6 O_5$	98.86	484.3377	15.58	161406	
Ganoderic acid F	$C_{32} H_{42} O_9$	98.65	570.2828	17.75	111046	

Table 4: List of abundant compounds in leaf methanolic extract of *S. multiflora* in positive mode.

Name	Formula	Score	Mass	RT	Abundance	Compound structure (only Abundance)
Terbinafine	$C_{21}H_{25}N$	52.62	291.2031	3.837	944358	
Kaempferol 4'-glucoside 7-rhamnoside	$C_{27}H_{30}O_{15}$	84.62	594.1552	4.704	94603	
Calendoflaside	$C_{28}H_{32}O_{15}$	83.09	608.1706	6.034	295390	
5-Oxoavermectin ''2a'' aglycone	$C_{34}H_{48}O_{9}$	51.92	599.3301	7.473	187098	
Pallidol 3-glucoside	$C_{34}H_{32}O_{11}$	72.02	616.199	7.476	77764	
Loganin pentaacetate	$C_{27}H_{36}O_{15}$	98.88	600.2046	7.712	996531	
LysoPE(22:5(4Z,7Z,10Z,13Z,16Z)/0:0)	$C_{27}H_{46}NO_{7}P$	55.55	527.3085	8.545	61008	
Cinncassiol C3	$C_{20}H_{30}O_{7}$	97.41	382.1995	9.589	108013	
8beta-Angeloyloxy-15-hydroxy-1alpha,10R-dimethoxy-3-oxo-11(13)-germacren-12,6alpha-olide	$C_{22}H_{32}O_{8}$	98.64	424.2099	10.009	233214	
16-Ketoestradiol	$C_{18}H_{22}O_{3}$	81.45	286.1577	10.046	144491	

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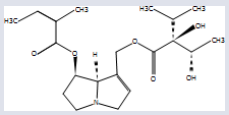
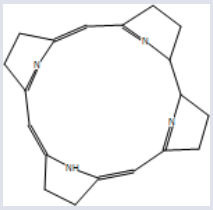
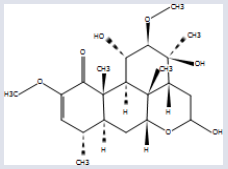
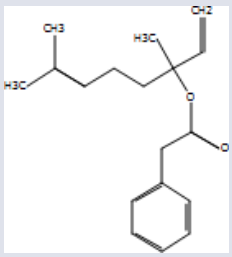
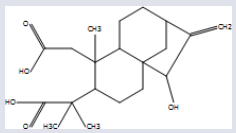
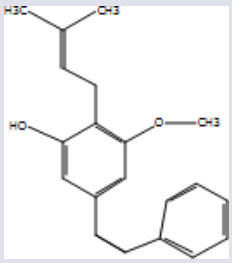
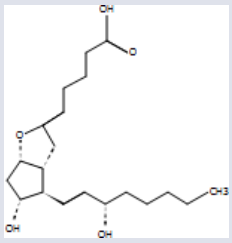
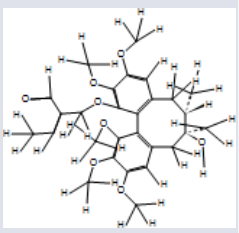
Prosolanapyrone II	$C_{18}H_{24}O_4$	83.78	304.1682	10.189	176297	
Symlandine	$C_{20}H_{31}NO_6$	51.53	381.2127	10.305	78973	
Corrinoid	$C_{19}H_{22}N_4$	83.82	306.1842	10.56	420262	
Nigakihemiacetal A	$C_{22}H_{34}O_7$	97.83	410.2307	11.304	216139	
Linalyl phenylacetate	$C_{18}H_{24}O_2$	82.07	272.1786	11.307	166412	
Oryzalic acid B	$C_{20}H_{30}O_5$	81.34	350.2102	11.345	162549	
Longistylin A	$C_{20}H_{22}O_2$	91.31	294.1607	11.632	263453	
Prostaglandin I2	$C_{20}H_{32}O_5$	97.84	352.2255	12.088	135209	
Tigloylgomicin H	$C_{28}H_{36}O_8$	85.8	500.2382	12.852	174418	

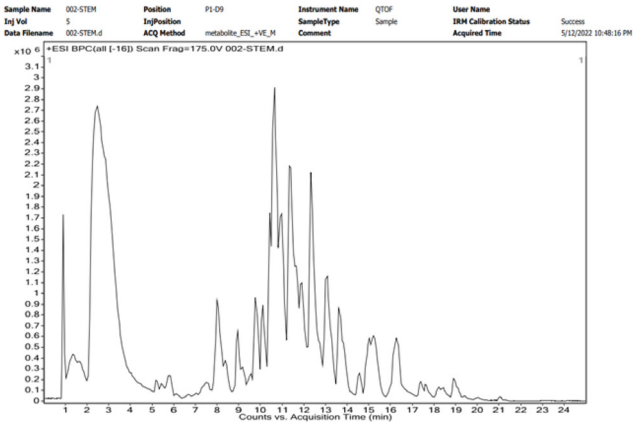


Figure 5: LCMS chromatogram of stem extract in positive mode.

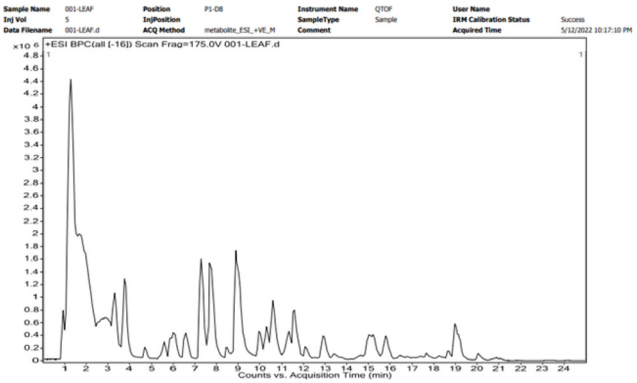


Figure 6: LCMS chromatogram of leaf extract in positive mode.

thereby making them physico-chemically fit. Qualitative phytochemical analysis was conducted on both the stem and leaf using different solvents based on polarity. It was observed that the methanolic extract could give more compounds in comparison with the other solvents used. Among these, the leaf methanolic extract resolved many compounds such as saponins, phenols, alkaloids, flavonoids etc. Alkaloids and terpenoids are potent hypoglycaemic, antihyperglycaemic, and glucose-suppressive agents by facilitating insulin release from beta-pancreatic cells, inhibiting glucose absorption in the gut, stimulating glycogenesis in the liver and increasing glucose utilization by the body.[4] In Wistar strain albino rats, extracts from *Eremurus himalaicus* were useful for establishing hypoglycaemic activity.[5] The physicochemical features reported from *Eclipta alba* and *Lippia nodiflora* were at par with the present data. Quinones, coumarins and flavonoids were totally absent in the extracts.[6]

Murniaty *et al.*[7] reported that Ethanol extract of *Clerodendrum fragrans* leaves contained many secondary metabolites including alkaloids, triterpenoids, saponins, tannins, flavonoids and quinones. According to Oladeji[8] medicinal plants and their phytocompounds are responsible for their therapeutic effectiveness. The polarity and type of solvent used during the extraction process have an influence on the medicinal properties due to difference in the solubility of the phytochemicals in the different solvents.[9] These results are similar to the report of phytochemicals conducted by Mohammed *et al.*[10] which showed the presence of flavonoids, steroids, alkaloids, and tannins in their chloroform, ethanol and aqueous extracts.

Flavonoids and terpenoids have been proven to be useful in preventing and treating many diseases. They are reported to possess antimicrobial, antiparasitic, anti-inflammatory, antifungal, antiviral and anti-allergenic properties, as well as in the management of cancer. They are also used in the regulation of immune systems.[11] From this study, it can be understood that *S. multiflora* extracts have various medicinal values due to the presence of a variety of phytochemicals. From validated scientific reports, the phytochemical compounds found in this plant have been reported to have beneficial health effects on humans.[12,13]

An effective technique to determine compounds accurately is HPTLC. By comparing the R_f values, peak area, and area percentage obtained from the fingerprinting to the reference standards, it is possible to identify unknown compounds. The concentration of the compounds may also be calculated from the peak area values. The HPTLC fingerprinting approach has been a feasible and perfect methodology for evaluating the quality of herbal medicine and its authentication.[14] According to Frommenwiler *et al.*[15] HPTLC fingerprinting data can be used for quantitative and qualitative determination of many phytochemicals. So the work carried out to unveil the variation in phytochemical constituents and their concentrations in *S. multiflora.* The HPTLC results of *Beta vulgaris* and *Bixa orellana* confirmed the presence of 16 different types of alkaloids bands and validated 11 different R_f values ranged from 0.03 to 0.93.[16] The qualitative analysis of methanolic extracts of leaf and root of *Hypochaeris radicata* using HPTLC confirmed the presence of secondary compounds like alkaloids, glycosides, saponins, flavonoids, and terpenoids. The HPTLC profiles show the medicinal importance which supports the traditional therapeutic uses of this species.[17] Using HPTLC and a single quadrupole MS, measured methylxanthines, phenolic compounds, and saponins in mate tea aqueous extracts.[18] Similarly, the profiling of *Salvia miltiorrhiza*, HPTLC-MS chromatographic settings included HPTLC silica gel plates with developing solvent

toluene-chloroform-ethyl acetate-methanol-formic acid (4:6:8:1:1) also supports the present data.

TLC plates visualized under white light and UV of wavelengths 254 nm and 366 nm revealed bands of separated compounds and substantiated the results. It was observed that the leaf methanolic extract could resolve many phytoconstituents at 366 nm. A wide range of bioactive compounds of medicinal significance has been reported from *Fumaria* species using HPTLC method. The HPTLC analysis of *Fumaria vaillantii* showed protopine and rutin in methanol extract with R_f values 0.51 and 0.26, respectively. Some of *Fumaria* species include bioactive phytochemicals such as alkaloids, polyphenols, and flavonoids, they have been shown to have antifungal, antibacterial, and anti-inflammatory properties.[19] Thus the present data of HPTLC from *S. multiflora* also invites its scope in drug industries.

The phytochemical fingerprint of methanolic stem and leaf extract from *S. multiflora* was estimated using LC-MS conditions. The experiments were carried out to identify the phytochemicals based on the intensity and retention values through LC-MS. Many of the compounds recorded in both stem and leaf were pharmaceutically proven molecules. Pharmacologically relevant compounds were abundant more in leaf extract as compared to that stem extract. It was found that many of the compounds such as corchorifatty acid, loganin, benazepril, nicotiflorin, vicenin 2, N-Nitrosofolic acid, flucetosulfuronetc were found to possess many pharmacological activities. It was profiled that the leaves of selected *Yucca* species with antimicrobial potentiality had 26 fatty acids with more saturated than unsaturated ones, 21 hydrocarbons constituting 39.64% of the unsaponifiable, while 3 sterols 42.36% were detected among which, major compound was γ-sitosterol.[20]

A total of 60 compounds, including 16 polyphenols, 43 flavonoids, and one quinic acid, were identified from *Morus alba* by using high-performance liquid chromatography.[21] LC–MS/MS analysis of the alcoholic extract of leaves of *Y. aloifolia* variegata L. in the positive mode proved to be better in the identification of saponins.[20] Around 11 secondary metabolites in traditional Korean medicine - Sogunjung decoction by LC-MS/MS MRM in positive and negative ion modes revealed compounds like gallate, magnoflorine, albiflorin, liquiritinapioside, liquiritin, liquiritigenin, paeoniflorin, coumarate, benzoylpaeoniflorin, cinnamaldehyde and glycyrrhizin.[22]

It was reported the correlation between phenolics versus antiradical power of elderberry flowers or fruits. Nine fractions were identified such as protocatechuic, 4-benzoic, caffeic, p-coumarate, salicylate, ferulate, rutin, isoquercetin and apigenin-7-glucoside.[23] Meanwhile, Sambucus flowers showed 15 polyphenols and additionally gallate, gentisic, vanillate, sinapate, kaempferol-3-rutinoside and astragalin. The ethanolic extract of *Phyllanthus* species was subjected to HPLC-ESI-QTOF-MS/MS and recorded 11 quercetin derivatives like quercetin 3-O-hexoside, rutin, quercetin 3-isorhamninoside, quercetin derivative, quercetin derivative, quercetin-di-O-hexoside, quercetin 3-sambubioside, quercetin-O-hexoside, quercetin 3-arabinoside, qauercetin-3,4-di-O-glucoside and quercetin 3-O-glucuronides; kaempferol 3-O-glucuronide, kaempferol- hexoside, 7 kaempferol derivatives - robinin, kaempferol-3-O-rutinoside, kaempferol-3-O- hexoside and kaempferol-O-hexoside.[24] In addition, quinic, ellagic acid-O-glucuronide, ellagic acid-O-arabinoside, caffeic, gallic, coumaric, methyl gallate,ethyl gallate, protocatechuic, ellagic acid-O-dihexoside, ellagic acid-O-hexoside, methy-O-ellagic acid, dimethyl-O-ellagic and trimethyl-O-ellagic acid.

LC-MS profile of *Punica granatum* peel extracts recorded the major component as valoneic acid dilactone, hexoside and coumaric acid.[25] Similarly quercetin 3-O-glucoside, llarein 7-O-pentoside, isoscute, and quercetin 3-O-pentoside were deduced from the seed extracts of *Strychnos potatorum*.[26] Secondary metabolites are screened and characterized diterpenes and flavones from the methanolic extract of *Andrographis paniculate* using LC-MS/MS.[27] A total of 74 fractions (25 from positive and 49 from negative ion mode) were recorded. Andrographolide (15%) was highest in positive and negative ion modes. All these works substantiate the phytochemicals recorded from *S. multiflora* were unique and pharmaceutically important.

CONCLUSION

Drug designing requires the isolation and identification of phyto-molecules having pharmacological potentiality. The results of the present study revealed the presence of unique phytochemicals in the methanolic extracts of *Suregada multiflora* stem and leaf, which may justify its therapeutical value in curing many ailments. The HPTLC and LC-MS investigation was conducted for phytochemical profiling, and by comparing the R_f values, retention times, and mass values of the compounds with the reference standards, it assisted in the identification of bio-lead compounds. Future studies are warranted to isolate the novel molecule and evaluate its anti-inflammatory potential.

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CONFLICT OF INTEREST

The author declares that there is no conflict of interest.

ABBREVIATIONS

ATS4: Automatic TLC Sampler 4; **HPTLC:** High-Performance Thin Layer Chromatography; **HRLCMSMS-qTOF:** High-Resolution Liquid Chromatography Mass Spectrometer Quadrupole Time-of-Flight; **LC-MS:** Liquid chromatography-mass spectrometry; **R_f value:** Retardation factor value; **SDBS:** Spectral Database for Organic Compounds.

SUMMARY

From the present study, it can be summarized that *Suregada multiflora* has a plethora of secondary metabolites, and can be used in developing crude drugs or formulating medications from different parts of *S. multiflora* against various ailments. Further *in vitro* and *in vivo* studies are needed to develop it as a crude drug.

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