



PHARMACOGNOSTICAL AND PHYTOCHEMICAL SCREENING OF HEMIGRAPHIS COLORATA BLUME

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ABSTRACT

The folk medicine system uses the plant products for the treatment of numerous diseases. Studies by various researchers have proved that plants are one of the major sources for drug. The curative properties of medicinal plants are due to the presence of various chemical substances of different composition which occur as secondary metabolites. Extraction of leaves of Hemigraphis Colorata Blume using various solvents by using Soxhlet apparatus. To determine the structural formula of various compounds isolated from Hemigraphis Colorata Blume. Phytochemical screening of crude drug extracts. The present work revealed that the phytochemical analysis of ethanolic extract of Hemigraphis colorata contain carbohydrate, protein, flavonoid, saponin, tannin, steroid and glycoside. The chloroform extract shows the presence of carbohydrate, protein, aminoacid, alkaloid, tannin, steroid and glycoside. The benzene extract indicate the presence of tannin, terpenoid, gum, carbohydrate, protein, steroid and glycoside. The moisture content in dry leaves of Hemigraphis Colorata Blume is 4.7%, total ash value (12.6%). The physicochemical parameters such as ash value and determination of moisture loss were used to determine the quality and purity of a crude drug. The biochemical analysis of Hemigraphis Colorata Blume revealed that of carbohydrate, of protein and others. Hemigraphis Colorata Blume possessed considerable level of bioactive compounds and therefor, these species can be used as a potential source of drugs. The plant has immense power to cure fresh wound, ulcers, inflammations and skin complaints.

Key words: Pharmacognostical Phytochemical Colorata Blume Hemigraphis.

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INTRODUCTION

At the present, the cutting edge customary medical services is troubled with extraordinary issues of perilous medications, persistent illnesses, safe contaminations, immune system problems and degenerative issues of maturing notwithstanding of incredible advances. Current allopathic framework has created many modern and expensive demonstrative techniques which at the occasions have made it very extravagant and past the span of average person.

Numerous cutting edge manufactured medications may hurt more than they help in relieving infections by its genuine results (Anitha VT, *et al* 2012). On opposite, customary medications which utilize plants are substantially more regarded being more protected without destructive impacts and nearly more affordable than numerous allopathic prescriptions (Karpagam, *et al.* 2008) Without a doubt, the plant realm actually holds numerous types of plants containing substances of therapeutic worth which presently can't seem to be found. Characteristic items particularly which are gotten from plants have been utilized as a hotspot for different restorative cycles for long occasions from the human progress (Akhil, *et al* 2013). The most well-known system of medication advancement from plants is cautious perception of utilization of normal assets in people medication in various societies by ethanopharmacology. In the new pattern, natural medications are recommended

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Quick Response code



Received: 25.09.2021

Revised: 12.10.2021

Accepted: 22.11.2021

plentifully in present day treatment in light of the extraordinary arrangement of security and less poisonousness profiles. Numerous significant medications utilized in medication today are straightforwardly or in a roundabout way got from plants because of their bioactive constituents, for example, alkaloids, steroids, tannins, saponins, glycosides, unstable oil. There are numerous photochemical in spices and each works in an unexpected way (Bhardwaj, *et al.* 2012). These phytochemical have different medical advantages, for example, cell reinforcement, antimicrobial, calming, malignancy preventive, antidiabetic and antihypertensive impacts, and so forth. By definition, conventional utilization of natural drugs infers considerable chronicled use, and this is surely valid for some items that are accessible as customary home grown medications. In many agricultural nations, an enormous extent of the populace depends on customary professionals and their armamentarium of restorative plants to meet medical services needs. In this advanced setting, fixings are now and again advertised for utilizes that were never examined in the conventional mending frameworks from which they arose (Biju, *et al.* 2015). A model is the utilization of ephedra for weight reduction or athletic execution upgrade.

The disclosure and advancement of effectual restorative specialist from characteristic sources gave persuading proof that plants could be a wellspring of novel medications. Western medication utilize numerous medications extricated from common items: atropine, cocaine, digitoxin, ephedrine, hyoscine, codeine, morphine, pilocarpine, quinine, reserpine, taxol, warfarin, menthol and so forth. While the regular item disengaged as the dynamic compound would not generally be reasonable for advancement as a compelling medication, it can give an appropriate lead to transformation into a clinically helpful specialist (Daniel, *et al.* 2012). In present day drug disclosure and advancement measures, characteristic items assume a significant job at the beginning phase of lead revelation, for example revelation of the dynamic (dictated by different bioassays) regular particle, which itself or its primary analogs could be an ideal medication up-and-comer. Spices and spice based treatment have been utilized by different social orders all around the world for the treatment of numerous illnesses has been by and by for quite a while. Throughout the long term, an assortment of therapeutic plants have been exceptionally well known for the fix of various both human and creature infections (Devi, *et al.* 2013). The poisonousness profile of the advanced medication which can prompt extreme dangerous obsessive condition requests an incredible clinical consideration. In ongoing patterns scientists are attempting to disengage the dynamic phytoconstituents from restoratively potential plants. As these dynamic phytoconstituents viably follow up on the objective site they are viewed as equipotent to manufactured medications. As referenced above, there are

countless plants in this world, and in which the greater part of them are going through numerous investigations and a large number of them are definitely not (Eleanor, *et al.* 2014). Be that as it may, lamentably we can't understand the vast majority of the intense therapeutic plants and their exercises. Subsequently, I have picked the plant *Hemigraphis alternata* (Burm. f.) T. Anders, (syn: *Hemigraphis colorata*) commonly known as Murikooti in Malayalam for my postulation work, to uncover its therapeutic incentive by demonstrating experimentally.

MATERIALS AND METHODS

Plant Collection

Hemigraphis colorata leaves were collected from Kerala. Chelembra gets about of rainfall high and situated at a height of approximately 163 ft. above sea level. Fresh leaves of the selected plant materials were collected during August (Gayathri, *et al.* 2012). The leaves were washed in running tap water. The leaves are shade dried and ground to fine powder and stored in air tight container for further analysis.

Preparation of extracts Pilot

Small powdered leaf was collected and 15g of it were measured and introduced into 100ml of Petroleum ether, Ethyl acetate, water & Ethanol. Extraction is carried out by shaker system for 48hrs. The nature and yield of the extract were noted. The extracts were stored in a refrigerator at 4°C for further studies.

Main

The leaves were dried at room temperature for 10 days. At that point these were processed into powder by mechanical processor. This powder was successively separated to their expanding extremity with Petroleum ether, Ethyl acetate, water & Ethanol. About 500g of powdered leaf was consistently pressed into a thimble in a Soxhlet apparatus and fractionated with 1000ml of Petroleum ether, Ethyl acetate, Ethanol and water separately. Consistent warmth was given by Mantox radiator to reusing of the dissolvable. The cycle of extraction proceeds for 1-2 hours for every solvents. The abundance dissolvable was vanished and the dried extract was kept in cooler at 4°C for their future use in phytochemical examination and pharmacological screenings.

Preliminary Phytochemical Analysis

Test for Carbohydrates

To 2 ml test solution add 2 drops of the Molisch reagent. The solution is poured slowly into test tube containing 2ml of concentrated sulphuric acid. So that two layers form.

Test for Protein

It is used to determine the presence of peptide bonds in protein. To 3ml of test sample add 3% sodium hydroxide and few drops of 1% copper sulphate

Test for Starch

Mix 3ml test solutions. A few drops of dilute iodine solutions. Blue colour appear. It disappears on boiling and reappears on cooling.

Test for Steroids

To 2ml of extract add 2ml chloroform & add 2ml concentrate sulphuric acid. Shake well; chloroform layer appear red

Test for amino acid

To 5ml of test sample solution add a few drops of 40% sodium hydroxide & 10% lead acetate boiled.

Test for Glycosides

To the extract add Glacial acetic acid, few drops of 5% ferric chloride and concentrated sulphuric acid are added and observed for a reddish brown coloration at

Test for Flavonoid

To 2ml of extract add few drops of ammonia solution.

Test for Alkaloid

To 0.5g of each extract add 5ml of 1% aqueous hydrochloric acid and kept in water bath, 1ml of filtrate is to be treated with Mayer's reagent.

Test for Tannin

To 0.5ml of extract 1ml of water and 1-2 drops of ferric chloride solution was added.

Test for Saponins

To 1ml extract add 2ml distilled water and shake it persistent

Test for Terpenoid

2ml of extract was mixed with 2ml chloroform in a test tube. To this 3ml concentrated sulphuric acid was carefully added along the wall of the test tube,

Test for Gums

To 1ml of extract add 3ml of dil. hydrochloric acid, fehling's solution is added drop by drop, till red coloration.

Thin layer chromatographic profiling

TLC is perhaps the easiest and greatest often applied method for assessing the purity of different extracts. In TLC the mobile phase is a thin layer of adsorbent fast over a sheet of a glass or plastic. Calcium

sulphate for an organic polymer serves to bind the adsorbent to the sheet. A small quantity of sample is placed near the bottom of solvent; the coldness to which the solvent moves the compound up the chromatogram. The sheet is depending on the ability of the multiple to adhere to this adsorbent system. More often than not adsorbent system can be found to separate greatest component of given mixture (Alhassan, *et al.* 2012). As a rule adsorbent framework can be found to isolate most part of given blend. This method is helpful for intensifies that are heat delicate or non-volatile.

- Practical requirements
- Stationary phases
- Glass plates
- Mobile phase
- Preparation and activation of TLC plates
- Application of sample
- Development tank
- Development technique
- Detecting or visualizing agents

A Glass Plates

Specific dimensions like 20 cm 20cm (full plate), 20cm 10cm (half plate), 20cm*5cm (quarter plate) can be used. It should be good quality and can withstand temperature used for drying.

Preparation and activation of TLC plates

Mixture of stationary phase and water forms slurry. TLC plates prepared by pouring, dipping, spraying or spreading. In pouring technique, slurry prepared and poured on the glass plate. Slurry spread uniformly on glass plate. Plates are dried in oven. In dipping technique two plates dipped in slurry, separated and dried. Disadvantage is that large quantity slurry required.

Spreading technique

In this technique TLC spreader is used. The glass plates are stacked on base plate. Slurry poured in the reservoir of TLC spreader. Thickness adjusted using knob in the spreader. 0.25 mm is normally used thickness for analytical purposes. Spreader rolled on the plates and air drying the plates. Plates are activated by keeping at 100 degree Celsius to 120 degree Celsius for one hour. Activated plates can be stored in thermostatically controlled oven for further use.

Application of sample

The concentration of sample of standard should be minimum for good spots. 2- 5ug of 1% solution of standard or sample spotted using capillary tubes. Spots should be 2 cm above the base of plate and spotting area should not be immersed in mobile phase. At least 4 spots can be spotted on quarter plate.

Development tank

The developing tanks require more solvent for developing tank –having hump in the middle and require less solvent. It should be lined inside with filter paper moistened with mobile phase

Mobile phase

The solvent or mobile phase depends on various factors.

- Nature of substance
- Nature of stationary phase
- Mode of chromatography
- Separation to be achieved. Analytical or preparative

PROCEDURE

Commercial sheet pre coated with alumina or silica gel are available. Select a solvent by testing out the samples in various solvents. Dissolve a small sample of the unknown in different flask containing solvents of different polarity. Place the TLC plates, the spotted side down in to the chamber so that the lower the pencil line about the solvents(Kokate, *et al.* 2001). Remove the plate from the development chamber and allowed to dry. Plate is placed under UV light, dark spots are observed.

Quantitative analysis

$$R_f = \frac{\text{Distance moved by solute}}{\text{Distance moved by solvent}}$$

R_f = retardation factor

Procedure for the LC-MS analysis

LC-MS has become strategy for decision in numerous stages during drug improvement measure by utilizing distinctive dissolvable concentrates. Ongoing advances incorporates electro splash, thermo shower, and particle shower ionization strategies which offer exceptional benefits of high location affectability and explicitness, fluid optional particle mass spectroscopy, later laser mass spectroscopy with 600 MHz offers precise assurance of atomic weight proteins, peptides. Isotopes example can be recognized by this procedure. For the latest LC-MS available, a programmed technique is remembered for the product bundle to tune and adjust in the ESI mode(Lim, *et al.* 2009). In any case, more established instruments or potentially unmistakable applications actually require manual or self-loader systems to enhance the boundaries that influence particle location. In a LC-MS instrument, the mass spectrometer is tuned and adjusted in three steps:(1) particle source and transmission streamlining, (2) MS alignment, and (3) calibrating. According to the above determined standard technique based to set particulars, for example, LC Column: Reverse Phase C-18, Pump: SPD 10 AVP, Mobile Phase: Water: Methanol: THF (50:40:10), Ionization Mode: Electronic Spray Ionization, Mode:

Both Positive, Injection Volume: 10 Micro liter, Flow rate: 1.5 ml/min, Column temperature: 250c, Column: PHENOMENEX RP 18, Column measurement: 25 cm x2.5 mm, LC Detection: 264 nm, M/Z Range: 50-1000 and Soft product: Class V P Integrated and Library: Metwin 2.0

PASS (Prediction of activity spectra of substances)

PASS is the PC created program which gives the synchronous forecast of a few many organic action types for any medication like mixtures. In this bio informatics programming forecast depends on the investigation of design movement connections of (SAR) the preparation set including in excess of 30000 known organically dynamic mixtures(Ramnivas Rangheetha, *et al.* 2016). In this paper principally research the effect on the precision of anticipating the kinds of action with PASS by (a) decrease of the quantity of designs in the preparation set and (b) decrease of the quantity of known exercises in the preparation set. Here chiefly show that forecasts are hearty in spite of the avoidance of up to 60% of data .The mixtures from the MDDR data set are utilized to make heterogeneous preparing and assessment sets . To assess the movement range for another compound (C) its MNA descriptors are produced. For each kind of movement (j) the estimation of t_jC is determined. The probabilities of quality P_{aj} and nonappearance P_{ij} of j-th movement type in the compound are determined by the following conditions $A_j(P_a) \rightarrow t_jC$; $I_j(P_i) \rightarrow t_jC$ at the end of the day, P_a and P_i are the probabilities of having a place with the classes of dynamic and idle mixtures, individually. The aftereffect of forecast for another compound is the movement range, which is the positioned rundown of action types with assessed P_a and P_i esteems(. The positioning is executed on dropping request of P_a - P_i ; along these lines, more plausible movement types are at the highest point of anticipated range. Compound is considered as dynamic if P_a - P_i surpasses the cut off esteem 55. Naturally here utilize cut off of $P_a - P_i$ 0, however any client may acknowledge his own cut off esteem. Essential components of PASS incorporate the accompanying:

The current rendition of PASS contrasts basically from the past .

Biological Activity

Organic exercises in PASS are portrayed subjectively: presence or nonattendance. Rundown of action types that have been found for each compound addresses the natural action information in the preparation set. This rundown for current adaptation of PASS is accessible through Internet.

Synthetic Structure Description

In our paper distributed as of late we portrayed the foundation descriptors called "Staggered

Neighborhoods of Atoms" (MNA). MNA descriptors depend on structure portrayal, which doesn't indicate the bond types and incorporates hydrogen's as per valence and halfway charge of iotas. MNA descriptors are created as a recursively characterized grouping: 1. Zero level MNA descriptor for every particle is simply the imprint An of the molecule and 2. any next-level MNA descriptor for every molecule is the foundation documentation A(D1D2. . . Di. . .), where Di is the past level MNA descriptor for the I-th prompt neighbors of the molecule.

Preparing Set

The forecast depends on the examination of the preparation set of organically dynamic mixtures. For each compound from the preparation set here store MNA descriptors and a rundown of action types. Each special MNA descriptor is incorporated into the portrayed the current rendition of PASS the preparation set comprises of around 35000 naturally dynamic mixtures ordered from logical writing, in-house and business data sets. The descriptor's word reference contains around 36000 MNA descriptors (Saravanan, *et al.* 2010). In various distributed sources natural exercises are named by various terms. In PASS this data is addressed in a standard structure that joins all organic action information about identical mixtures gathered from numerous sources. The quantity of various sorts of movement surpasses 800, yet a significant number of them are addressed by under 6 mixtures. Absolute "movement range", i.e., the rundown of unsurprising sorts of natural action, incorporates in excess of 500 things descriptors word reference.

Forecast Procedure

To gauge the action range for another compound (C) its MNA descriptors are produced. For each sort of action (j) the estimation of t_jC is determined. The probabilities of quality P_{aj} and nonappearance P_{ij} of j-th action type in the compound are determined by the following conditions $A_j(P_a) = t_jC$; $I_j(P_i) = t_jC$ as such, P_a and P_i are the probabilities of having a place with the classes of dynamic and idle mixtures, Separately. The consequence of forecast for another compound is the movement range, which is the positioned rundown of action types with assessed P_a and P_i esteems. The positioning is executed on slipping request of $P_a - P_i$; in this way, more plausible action types are at the highest point of anticipated range. Compound is considered as dynamic if $P_a - P_i$ surpasses the cut-off esteem. Of course here utilize cut-off of $(P_a - P_i) > 0$, yet any client may acknowledge his own cut-off esteem.

RESULTS AND DISCUSSION

Plant collection

We were collected 5kg plant material and authenticated by botanist. Collected materials were dried

in a shade and remove the moisture, taken the weight 4.5 kg. Made powdered and taken the weight 4.25 Kg

Extract and their yield

Product yield was found to be 76.6% and which are utilized for pharmacological phytochemical and analytical screenings

Preliminary phytochemical screening

a. Ethanolic Extract

The phytochemical studies results revealed that the Molisch's test presence of brown color ring observation indicated the present of carbohydrates, by phosphomolybdic acid test Blue coloration of the spot indicated the presence of phenols. Shinoda test and Lead acetate test gave pink or red coloration of the solution indicated the presence of flavonoids Flocculent white precipitate also indicated the same. Formed dark blue or greenish grey coloration of the solution indicated the presence of tannins in the drug. Color changes was observed in the presence for steroids and dark pink or red coloration of the solution indicated the presence of terpenoids. Orange coloration of the spot indicated the presence of alkaloids. Yellow or reddish brown precipitation indicated the presence of alkaloids. Pink to red color solution indicates the presence of glycosides. Layer of foam formation indicates the present of Saponins. If the response to the test is indicated table-1 which indicates that the particular phytochemical constituents present in the extract.

b. Pet. ether extract

Results revealed that the Molisch's test presence of earthy colored shading ring perception demonstrated the present of starches, by phosphomolybdic analysis Blue coloration absent showed the shortfall of phenols. Shinoda test and Lead acetic acid derivation test gave pink or red shading of the arrangement showed the presence of flavonoids Flocculent white accelerate likewise demonstrated something very similar. Framed dull blue or greenish dim tinge of the arrangement demonstrated the presence of tannins in the medication. Shading changes were seen in the presence for steroids and dull pink or red tinge of the arrangement demonstrated the presence of terpenoids. Orange hue of the spot showed the presence of alkaloids. Yellow or rosy earthy colored precipitation demonstrated the presence of alkaloids. Pink to red shading arrangement shows the presence of glycosides. Layer of foam arrangement demonstrates the shortfall of Saponins. Violet shading missing so the shortfall of amino acids.

c. Ethyl Acetate

Various test reports are indicated that the Molisch's test occurrence of brown color ring observation indicated the presence of carbohydrates, by

phosphomolybdic acid test Blue coloration absence which expressed absence of phenols. Shinoda test and Lead acetate test gave pink or red coloration of the solution shown in the presence of flavonoids flocculent white precipitate also shown above same. Absence of Formed greenish grey coloration of the solution illustrated absent of tannins in the drug. Color changes were observed in the present for steroids and dark pink or red coloration of the solution shown the presence of terpenoids (Annapoorna, *et al.* 2013). Orange coloration of the spot shown the presence of alkaloids. Yellow or reddish brown precipitation illustrated in the presence of alkaloids. Pink to red color solution, presence of glycosides. Layer of foam formation indicates the absence of Saponins. Violet color was seen in the test tube in the present of amino acids

d. Water

The Molisch's test presence of natural hued concealing ring insight exhibited the present of starches, by phosphomolybdic fundamental investigation Blue hint missing showed the shortage of phenols. Shinoda test and Lead acidic corrosive determination test gave pink or red hint of the game plan showed the presence of flavonoids Flocculent white empower furthermore exhibited something comparative. Outlined faint blue or greenish faint concealing of the plan exhibited the presence of tannins in the drug. Concealing changes were found in the presence for steroids and faint pink or red shade of the game plan showed the presence of terpenoids. Orange concealing of the spot showed the presence of alkaloids. Yellow or bronzed hearty hued precipitation showed the presence of alkaloids. Pink to red concealing game plan shows the presence of glycosides. No layer of foam plan shows the deficiency of Saponins. Violet concealing present so the presence of amino acids.

Thin layer chromatographic profiling

Plan to selected Solvent System: n-Hexane : Ethyl Acetate (4:1). Extracts were given codes such as F1-Pet.ether, F2-Ethanol F3- Ethyl acetate and F4-Water. Second time spotted same extracts F5-Pet.ether,

F6-Ethanol and F7- Ethyl acetate. Rf value was founded values compared with standard values.

Procedure for the LC-MS analysis

- LCMS study Specifications:
- Lc Column: Reverse Phase C-18
- Pump: Spd 10 Avp
- Mobile Phase: Acetonitrile : Methanol: (50:50)
- Ionisation Mode: Electronic Spray Ionization
- Mode: Both Positive
- Injection Volume: 10 Micro Litre
- Flow Rate: 1.5 Ml/Min
- Column Temperature: 25⁰c
- Column: Phenomenex Rp 18
- Column Dimension: 25 Cm X2.5 Mm
- Lc Detection : 264 Nm
- M/Z Range: 50-1000
- Soft Ware: Class V P Integrated.
- Library: Metwin 2.0

We present a far reaching, delicate, and exceptionally explicit negative particle electrospray LC/MS strategy for recognizing all primary classes of same plant various concentrates from chosen bioactive plant introduced various pinnacles normal for steroids, flavone C-glycosides, and xanthenes. To show the sort of data recorded on-line, the LC/MS spectra of two polyphenols have been chosen. . As indicated by these most presumably a flavone C-glycoside subbed by three hydroxy and one methoxy gatherings(Subramoniam, *et al.* 2001). Two isomeric flavones happening in Gentianaceae, isoscoparin and swertiajaponin, fitted with such information.

Comparison of reference data used to confirm the main constituents present in different extracts of *Hemigraphis Colorata* Blume were found to be Kaempferol, Rhamnetin, Rhamnoxanthin, Quercetin and Luteolin.

Pass Study

Results revealed that the all extract have some biological interaction with receptors and also which more prominently used active structurally resembled compounds are present in all extracts.

Table 1: Preliminary pharmacognostical study

S.No.	Parameters	GV
		(% w/w)
1	Extractive Values	
a.	Petroleum ether	16.23
b.	Ethyl acetate	3.3
c.	Ethanol	20
d.	Water	13.63
2	Ash Values	
a.	Total Ash	6.35
1	Petroleum ether	6.33

2	Ethyl acetate	6.23
3	Ethanol	6.34
4	Water	6.00
b.	Acid insoluble Ash	2.54
1	Petroleum ether	2.57
2	Ethyl acetate	2.60
3	Ethanol	2.56
4	Water	3.10
c.	Water soluble Ash	1.43
1	Petroleum ether	1.43
2	Ethyl acetate	1.44
3	Ethanol	1.45
4	Water	1.48
d.	Sulphated Ash	2.13
1	Petroleum ether	2.4
2	Ethyl acetate	2.14
3	Ethanol	2.15
4	Water	2.16
3	Loss on Drying	0.89
1	Petroleum ether	0.89
2	Ethyl acetate	0.88
3	Ethanol	0.88
4	Water	0.77
4	Crude fiber content	10.2
1	Petroleum ether	10.2
2	Ethyl acetate	10.2
3	Ethanol	10.3
4	Water	10.4

Table 2: Results of ethanolic extract of Hemigraphis Colorata Blume

Class of compounds	Tests performed	Results
Carbohydrates	Molisch's test	+
	Fehling's test	
Phenols	Phosphomolybdic acid test	+
Flavonoids	Shinoda test	+
	Lead acetate test	
Tannins	Braemer's test	+
Alkaloids	Wagner's	+
	Mayer's	+
	Draggendorf's test	+
Glycosides	Legal's test	+
	Brontranger's test	+
		+
Saponins	Foam test	+
Sterols	Salkowski's test	+
Amino acids	Ninhydrin test	+
Terpenoids	Lieberman Burchardt test	+

+Present -Absence

Table 3: Results of Pet. ether extract of Hemigraphis Colorata Blume

Class of compounds	Tests performed	Results
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Carbohydrates	Molisch's test Fehling's test	+
Phenols	Phosphomolybdic acid test	-
Flavonoids	Shinoda test Lead acetate test	+
Tannins	Braemer's test	+
Alkaloids	Wagner's Mayer's Draggendorf's test	+
Glycosides	Legal's test Brontranger's test	+
Saponins	Foam test	+
Sterols	Salkowski's test	+
Amino acids	Ninhydrin test	-
Terpenoids	Lieberman Burchardt test	+

+Present -Absence

Table 4: Results of ethyl acetate extract of Hemigraphis Colorata Blume

Class of compounds	Tests performed	Results
Carbohydrates	Molisch's test Fehling's test	+
Phenols	Phosphomolybdic acid test	-
Flavonoids	Shinoda test Lead acetate test	+
Tannins	Braemer's test	-
Alkaloids	Wagner's Mayer's Draggendorf's test	+
Glycosides	Legal's test Brontranger's test	+
Saponins	Foam test	+
Sterols	Salkowski's test	+
Amino acids	Ninhydrin test	+
Terpenoids	Lieberman Burchardt test	+

+Present -Absence

Table 5: Results of water extract of Hemigraphis Colorata Blume

Class of compounds	Tests performed	Results
Carbohydrates	Molisch's test Fehling's test	+
Phenols	Phosphomolybdic acid test	-
Flavonoids	Shinoda test Lead acetate test	+
Tannins	Braemer's test	+
Alkaloids	Wagner's Mayer's Draggendorf's test	+
Glycosides	Legal's test Brontranger's test	+

		+
Saponins	Foam test	-
Sterols	Salkowski's test	+
Amino acids	Ninhydrin test	+
Terpenoids	Lieberman Burchardt test	+

+Present -Absence

Table 6: Thin layer chromatographic profiling

Extracts	State	Detection	Rf
Pet.ether	Light yellowish sticky mass	UV at 254 nm& at 366 nm	0.79
Ethanol	Light Brown sticky mass	UV at 254 nm& at 366 nm	0.75
Ethyl acetate	Light yellowish brown	UV at 254 nm& at 366 nm	0.82
Water	Dark brown & Sticky	UV at 254 nm& at 366 nm	0.65

Table 7: LCMS Obtained Results of different extracts of Hemigraphis Colorata Blume

SL NO	Compound Name	Molecular Mass
1	Kaempferol	270.25
2	Rhamnetin	316.27
3	Rhamnoxanthin	416.39
4	Quercetin	302.24
5	Luteolin	286.25

Figure 1: TLC For Various Extracts : Visualization At UV 254 Nm

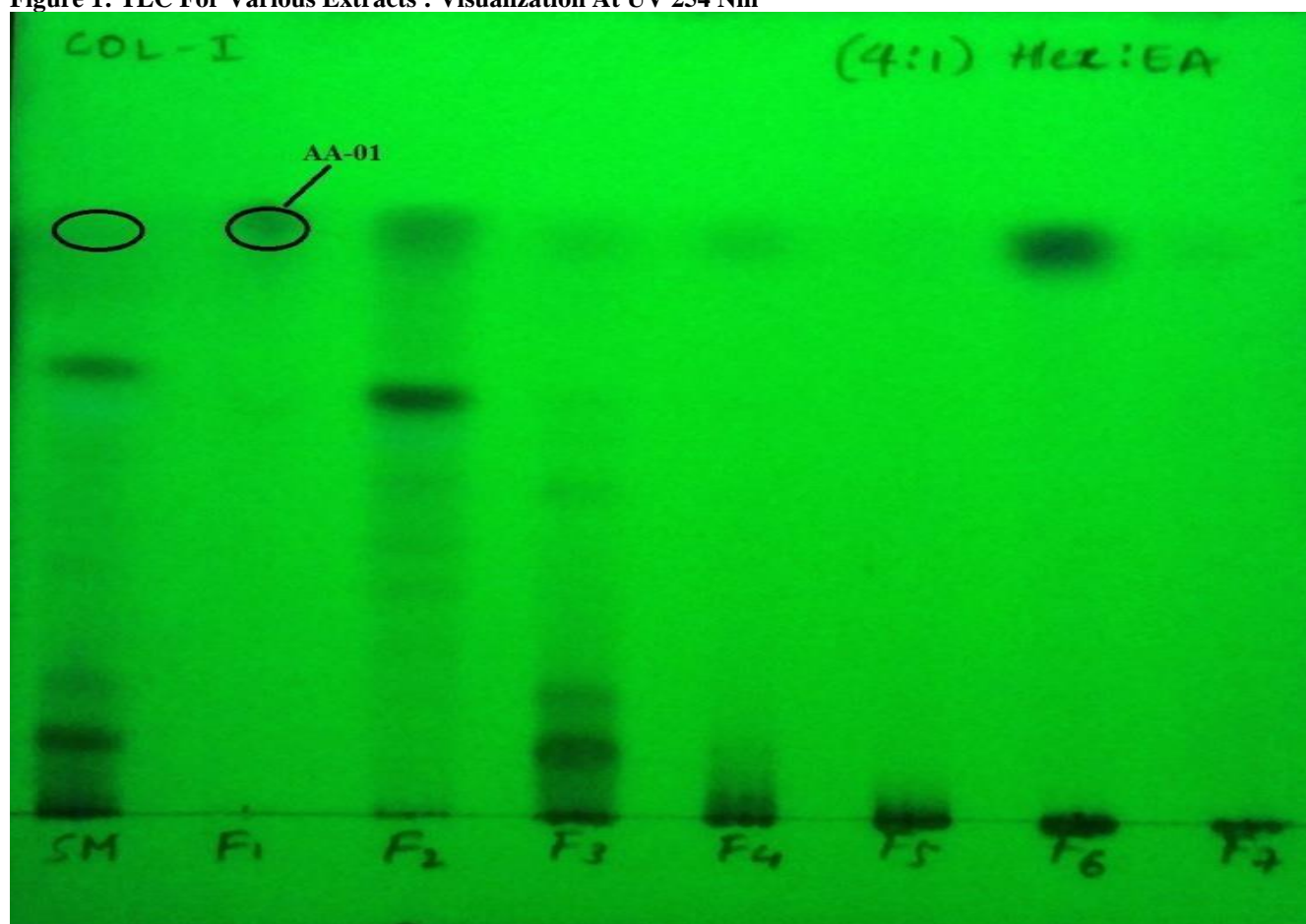


Fig.No:2: Visualization At UV 356 Nm

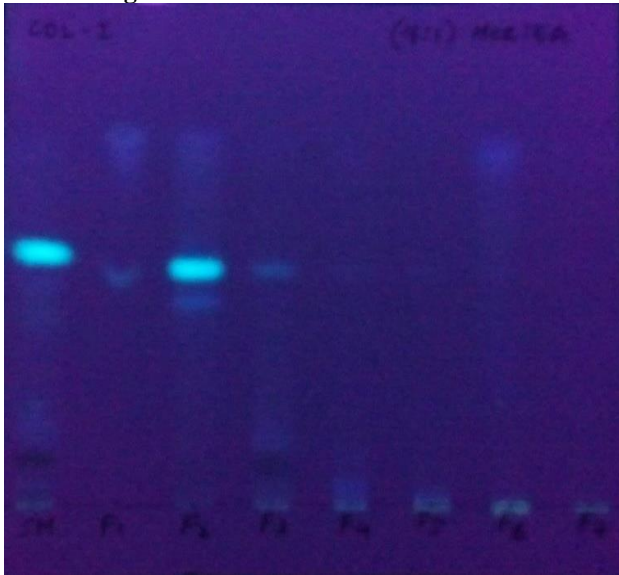


Fig.No:3: TLC Plate Normal

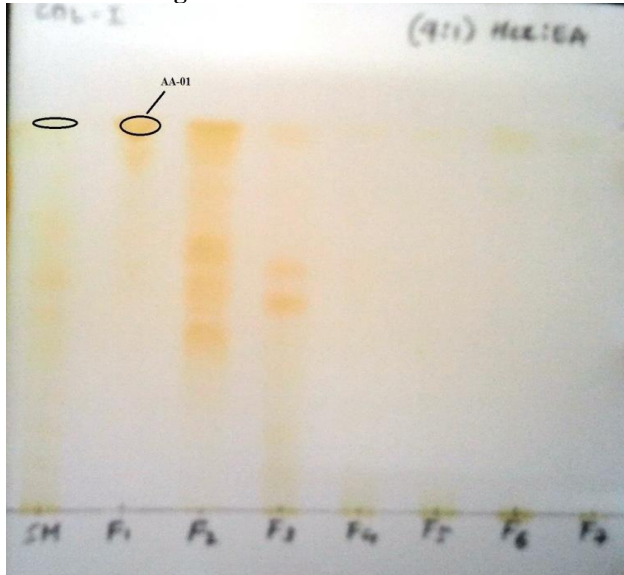


Figure 4: LCMS for Pet. Ether Extract

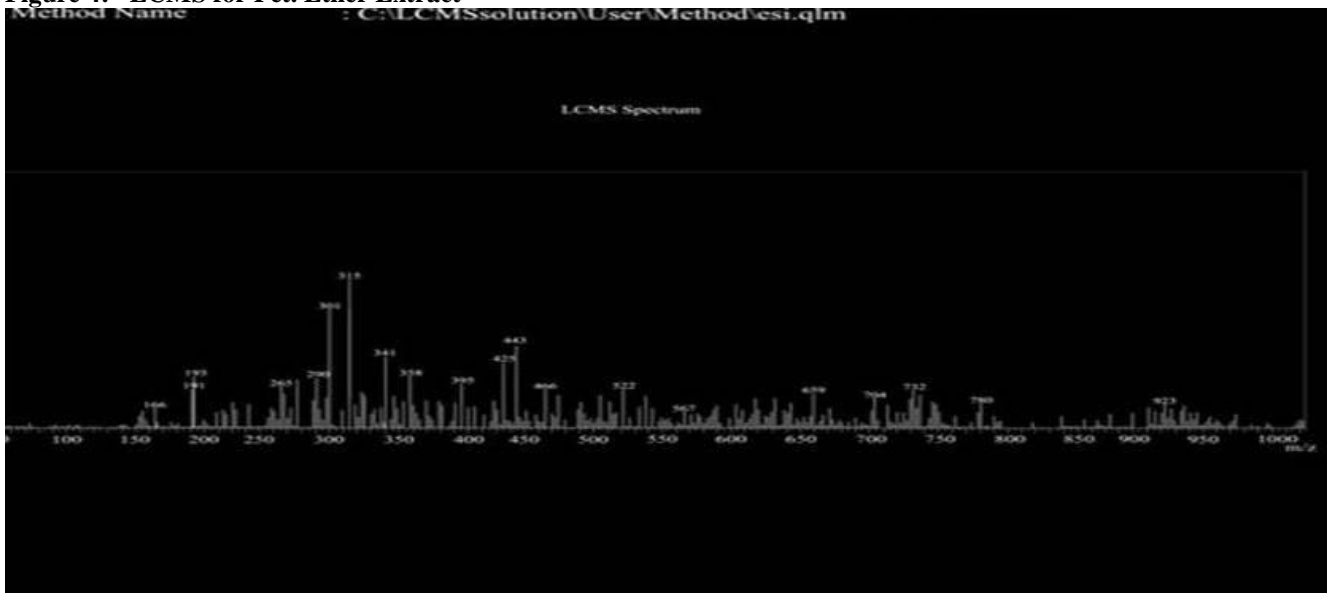


Figure 5: LCMS for Pet. Ethanollic Extract

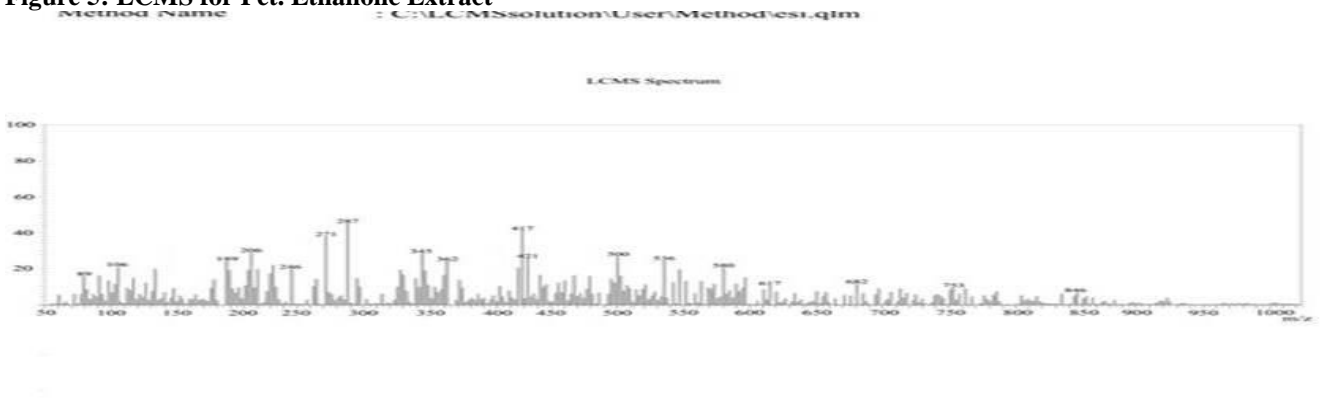
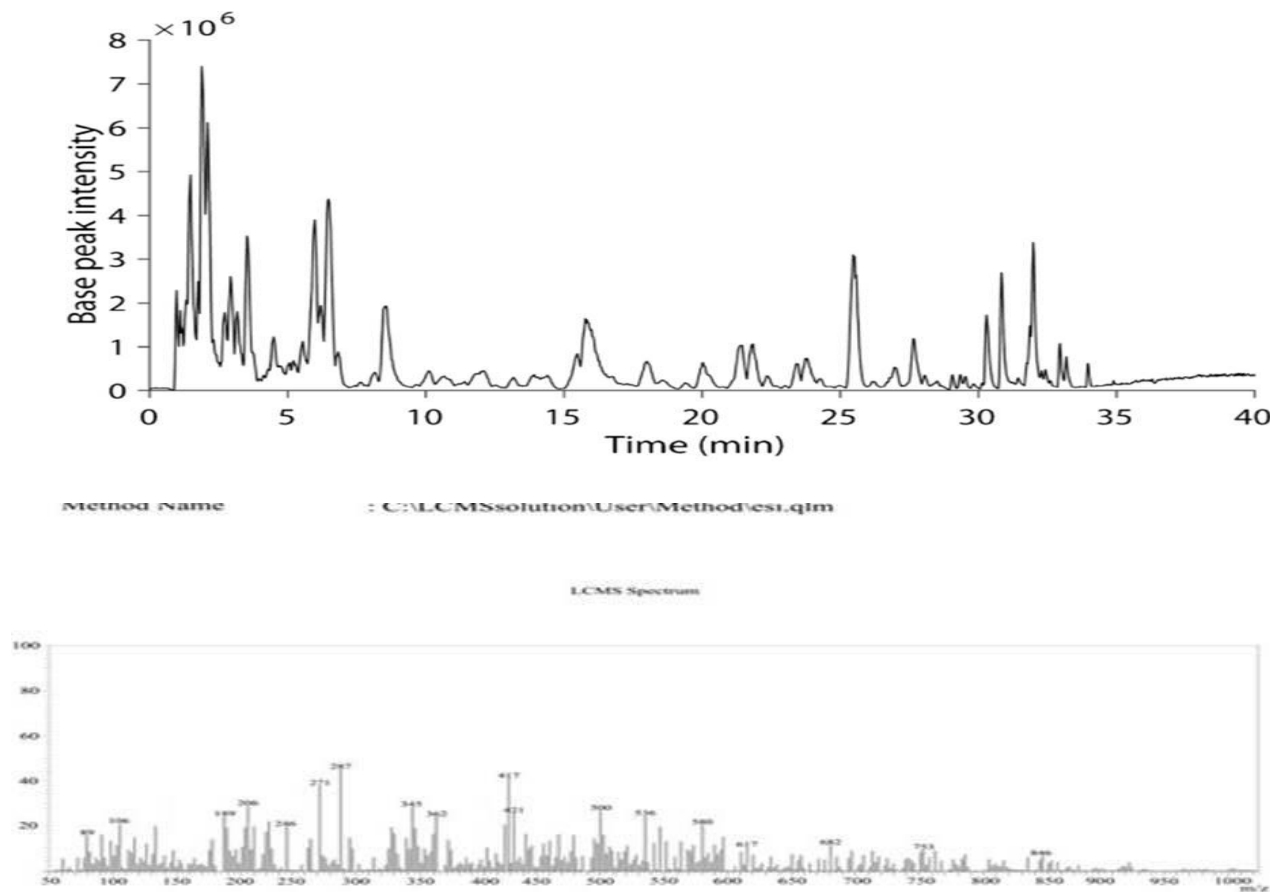


Figure 6: LCMS for ethyl acetate Extract



SUMMARY AND CONCLUSION

The traditional and folk medicine system uses the plant products for the treatment of various infectious diseases. The curative properties of medicinal plants are due to the presence of various chemical substances of different composition which occur as secondary metabolites (Arun, *et al.* 2013 & Adasivam, *et al.* 2008)

Hemigraphis colorata is traditional medicinal plant mainly used to treat cut and wounds. The present work revealed that the phytochemical analysis of ethanolic extract of Hemigraphis colorata contain carbohydrate,

protein, flavonoid, saponin, tannin, steroid and glycoside. The chloroform extract shows the presence of carbohydrate, protein, amino acid, alkaloid, tannin, steroid and glycoside (Asha, *et al.* 2014). The benzene extract indicate the presence of tannin, terpenoid, gum, carbohydrate, protein, steroid and glycoside. Hemigraphis colorata possessed considerable level of bioactive compounds and therefore, these species can be used as a potential source of drugs (Pulok, *et al.* 2006). The plant has immense power to cure fresh wound, ulcers, inflammations and skin complaints.

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