Available online www.jocpr.com

Journal of Chemical and Pharmaceutical Research, 2016, 8(5):614-617

Research Article

ISSN : 0975-7384
CODEN(USA) : JCPRC5

Preliminary Phytochemical and TLC Profiling of *Lantana camara* Leaf Extracts

Sunil H. Ganatra and Sevak B. Gurubaxani

Computational & Pharmaco-analytical Laboratory, Department of Chemistry, Institute of Science, Nagpur-440001, Maharashtra (India)

ABSTRACT

Lantana camara locally named as Ghaneri in Marathi is found in the gardens as an ornamental plant and at the roadside of most of the cities. This plant has high medicinal value as recognized by Indian traditional folk. It is known antimicrobial, fungicidal, insecticidal and immune-depressant. Lantana oil is popular against skin infections generally used for the cure and care of leprosy patients. So, in the present study an attempt has been made to chemically reinvestigate the bio-active components in leaf extracts of Lantana camara extracted sequentially in N-Hexame, ethyl acetate, acetone, methanol and ethanol solvents. The chemical investigation of these extracts show the presence of terpenes, saponins in all extracts. Steroids are reported in all extracts except methanol. Falavonoids are present in n-hexane and ethyl acetate extracts. Carbohydrates are present in n-hexane, ethyl acetate and acetone extract. Glycosides are present in acetone and methanol extracts. Coumarins class of compounds is reported in ethyl acetate, acetone and methanol. Tannins, Phlobatannins, Alkaloids, Proteins, Emodins, Anthraquinones, Anthocyanins and Leucoanthocyanins classes of compounds are absent in all extracts. The probable number of compounds in each extracts determined using TLC.

Keywords: Terpene, Saponin, Solvent Extract, Natural Product, TLC

INTRODUCTION

Lantana camara commonly known as Ghaneri (Marathi) and Raimuniya (Hindi) is a weed plant belonging to family *Verbenaceae* widely found in India. Traditional medicinal healers use the parts of this plant in the treatment of tetanus, epilepsy and gastrointestinal disorders etc. Their fresh roots are used by hill tribes for various types of dysentery. Powdered leaves are used for cuts, wounds, ulcers, and swellings. The concoction of its leaves is good for bilious fever, eczema and eruptions. The fruits of plant are given against tumor and rheumatism. Different parts of the plant show biological activities like hepatoprotective, wound healing, and anti-diabetic and are potential source for array of other such properties which are still not recognized due to the dearth of scientific studies [1, 2].

EXPERIMENTAL SECTION

Collection, Identification, Authentication of Plant and Sample Preparation

Plant was collected from outskirts of Gondia district of Vidharbha, Maharashtra; India. The plant parts needed for study were authenticated by Dr. (Mrs.) Rani V. Choubey, taxonomist and visiting faculty Department of Environmental Sciences, Institute of Science; Nagpur-440001.

The specimen was submitted to the herbarium of the department with authentication code, Research/Env./112. *Lantana camara* leaves were shed dried for one week and crushed in a home based grinder.

Cold extraction

For sequential extraction, 25 g. of fresh and dried plant materials were put in a brown glass container with 200 ml. solvent and allowed to stand at room temperature for a period of 3 days with short agitation in intervals until the soluble matter get dissolved. The extract thus obtained was decanted and filtered. The clear extract was subsequently concentrated using rotary vacuum evaporator. The same procedure is adopted in each case by taking same powder and a solvent with higher polarity. The hierarchy of solvent used was from non-polar solvents to polar solvents [3-5]. N-hexane, ethyl acetate, acetone, methanol and ethanol solvents were used for the extractions.

Preliminary phytochemical tests

Crude extracts evaluated for the presence of fifteen naturally occurring phytochemicals using standard methods as described by Harbone and Evans [6-12]. Chemicals used in these investigations were of AR grade procured from market and used without any further purification. The solvents are of spectroscopic grade and used without distillation. Phytochemical investigations were performed for all crude extract separately. The chemical methods used to identify phytochemicals are as follows:

Tannins: 1ml. of 5% ferric chloride to solvent free extract was added. Formation of bluish black or greenish black precipitate indicated the presence of tannin.

Flavonoids: To 1 ml. of extract, 10% of 1ml. $Pb(OAc)_4$ was added, an intense yellow colour was produced in the plant extract, which indicated the presence of flavonoids.

Terpenoids: 2 ml. of extract was mixed with 2 ml. of acetic anhydride. Then 2-3 drops of concentrated H_2SO_4 was added. A reddish brown precipitate colour at the interface formed indicated the presence of terpenoids.

Saponins: To a 5 ml. of extract, few drops of olive oil were added. The formation of frothy emulsion indicated the presence of saponins.

Steroid: 1ml. of extract dissolved in 10 ml. chloroform and equal volume of concentrated H_2SO_4 added by sides of test tube. The red colour of upper layer and yellow colour with green fluorescence in acid layer indicates the presence of steroids.

Phlobatannins: To 2 ml. of extract and 2 ml. of 1% HCl was added then the content was heated in a boiling test tube, formation of red precipitate indicated presence of Phlobatannins.

Carbohydrates: 2 ml. of extract was added with few drops of alcoholic alpha napthol, then 2ml of concentrated sulfuric acid was added slowly through the sides of the test tube, a purple to violet color ring appeared at the junction indicated the presence of carbohydrates.

Glycosides: 5 ml. of extract was treated with 2 ml. of chloroform and 2 ml. of glacial acetic acid, a violet to blue to green colouration indicated presence of glycosides.

Coumarins: To 2 ml. of extract, 3 ml. of 10% NaOH was added, a yellow coluration indicated presence of coumarins.

Alkaloids: A yellow precipitate by addition of Hager's reagent to 2 ml. of extract indicated presence of alkaloids.

Proteins: To 1 ml. of extract, when 1 ml. of sulphuric acid was added, a white precipitate indicated presence of proteins.

Emodins: 2 ml. of extract was added with 2 ml. of ammonium hydroxide and 3 ml. of benzene gave red colouration, indicated the presence of emodins.

Anthraquinones: To a 3 ml. of extract when 3 ml of benzene and 5 ml. of 10% ammonia solution was added a pink, violet or red colouration in ammonical layer indicated the presence of anthraquinones.

Anthocyanins: When 2 ml of extract was added with 2 ml. of 2N HCl and ammonia a pinkish red to bluish violet colour indicated the presence of Anthocyanins.

Leucoanthocyanins: Organic layer turns into red when isoamyl alcohol was added to 5 ml. of extract which indicated the presence of Leucoanthocyanins. The observations for phytochemical tests are shown in **table 1**.

Table 1. Preliminary phytochemical analysis of extracts of *L. camara* in different solvents by cold extraction method.

Phytochemicals	n-Hexane	Ethyl acetate	Acetone	Methanol	Ethanol
Tannins	--	--	--	--	--
Flavonoids	++	++	--	--	--
Terpenoids	++	++	++	++	++
Saponins	++	++	++	++	++
Steroids	++	++	++	--	++
Phlobatannins	--	--	--	--	--
Carbohydrates	++	++	++	--	--
Glycosides	--	--	++	++	--
Coumarins	--	++	++	++	--
Alkaloids	--	--	--	--	--
Proteins	--	--	--	--	--
Emodins	--	--	--	--	--
Anthraquinones	--	--	--	--	--
Anthocyanins	--	--	--	--	--
Leucoanthocyanins	--	--	--	--	--

Positive (++), Negative (--)

Thin layer chromatography

To ascertain the presence of various secondary bioactive components as reported in chemical analysis, extracts were further examined using thin layer chromatography (TLC). TLC were performed for various extract on aluminium plates with MERCK silica gel 60 F_{554} 0.25mm thick as stationary phase. These plates were developed in two different sets of mobile phases as described in literatures [8, 13-16] and their compositions are listed in **table 2.** The spots were visualized by exposing plates to Iodine vapour as described in various literatures [13-15].

Table 2. Mobile phases and their composition used during TLC [16].

Phase Number	Mobile Phase	Composition
I.	n-Hexane : Chloroform	9:1
II.	Ethyl acetate : Chloroform	9:1

Figure 1 shows the TLC photographs and table 3 shows the calculated R_f values for all studied TLC systems

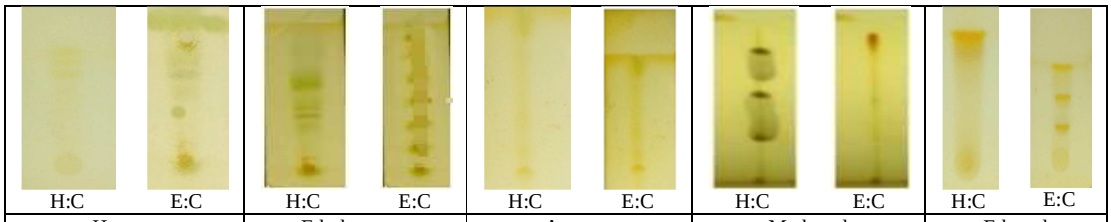

Figure 1. Photographs of TLC analysis using two different mobile phases

Table 3. Calculated R_f values for different mobile phases

Plant Extract	R_f Values			
	No. of spots in mobile phase I	Mobile Phase I (H : C)	No. of spots in mobile phase II	Mobile Phase II (E : C)
Hexane	3 spots/ tailing	0.6, 0.63, 0.65	3 spots	0.45, 0.63, 0.65
Ethyl acetate	5 spots/ tailing	0.33,0.42, 0.46,0.52, 0.56	6 spots	0.26, 0.33,0.54, 0.66, 0.78,0.80
Acetone	Tailing	----------	Tailing	----------
Methanol	3 spots/taing	0.36, 0.42, 0.46	3 spots	0.24, 0.36, 0.52
Ethanol	Ttailing	----------	3 spots	0.33, 0.46, 0.64

RESULTS

Test observations, as tabulated in table **2,** shows the presence of various secondary metabolites dissolved in various leaf extracts of *L. camara*. These investigations revealed the presence of terpenonids and saponins in all extracts. Steroids are reported in all extracts except methanol. Falavonoids are present in n-hexane and ethyl acetate extracts. Carbohydrates are present in n-hexane, ethyl acetate and acetone extract. Glycosides are present in acetone and methanol extracts. Coumarins class of compounds is reported in ethyl acetate, acetone and methanol. Tannins, Phlobatannins, Alkaloids , Proteins, Emodins, Anthraquinones, Anthocyanins and Leucoanthocyanins classes of compounds are absent in all extracts. The probable number of compounds in each extracts determined using TLC as shown in figure 1 and table 3. Highest number of spots reported in ethyl acetate extract. 5 spots in mobile phase I and 6 spots in mobile phase II. All other extracts shows atleast 3 spots in both mobile phases (I and II) except aceton extracts which shows tailing effect.

DISCUSSION

It was understood from the present study that the extracts of *L. camara* contain many phytochemicals as revealed by chemical analysis of extract and TLC analysis. This study provides the possible phytochemcials in *L. camara*. The finding of phytochemicals shows a way to perform detail investigations using GC-MS and LC-MS, which will confirm the presence of reported phytochemicals in our study.

CONCLUSION

The results obtained have shown the presence of terpenes and saponins classes of compounds. Along with them flavonoids, steroids, carbohydrates and coumarins are also reported. This study indicates that *L. camera* leaves has medicinally useful phytochemicals. The presence of terpenes in all extract indicates its confirm presence. Terpenes are good anti-cancer agents and hence further analysis of these extract by GC-MS followed by Column chromatography will provide the way to isolate terpenes from these plants.

Acknowledgement
R.T.M. Nagpur University for providing necessary scholarship to Shri Sevak Gurubaxani for his Ph.D. Research.

REFERENCES

[1] MK Lonare et al., *International Journal of Pharmaceutical Sciences and Research,* **2012,** 3(9): 3031-3035.
[2] SB Gurubaxani et al., *International Journal of Life Sciences*, Sp. Issue, **2016,** A6:137-140.
[3] M Ramya et al., *International Journal of Pharma and Bio Sciences*, **2013,** 4(1): 81-90.
[4] Akumu Edwin, Anthoney Swamy T et al., *International Journal of Pharmacy and Biological Sciences*, **2014,** 4(2): 28-34.
[5] S Handa et al., International centre for science and high technology, Trieste, Italy, **2008,** 21-25.
[6] JB Harbone, Phytochemical Methods: A guide to modern technique of plant analysis. 2nd ed., Chapman and Hall, UK, **1984.** 192-201.
[7] WC Evans, Trease and Evans' Pharmacognosy. 16th ed., Elsevier Ltd., London, **2009.**
[8] SH Ganatra et al., *International Journal of Pharmaceutical Sciences and Research,* **2013,** 4, 1135-1139.
[9] SH Ganatra et al., *Journal of Chemical and Pharmaceutical Research*, **2012,** 4(5), 2380-2384.
[10] D Kealey, Experiments in modern analytical chemistry, Springer science and Business Media, New York, **1986,** 42-44.
[11] A Matuszewska; A John, Acta Chromatographica, **2004**, 14.
[12] RV Malbasa, Acta Periodica Technologica, **2004,** 35, 199-205.
[13] M Medic-Saric, Croatica Chemica Acta, **2004,** 77(1-2): 361-366.
[14] SH Ganatra; AM Ramteke et. al., *Research Journal of Pharmacognosy and Phytochemistry*, **2012,** 4(4): 220-222.
[15] H Wagner; S Bladt, Plant Drug Analysis: A Thin Layer Chromatography, 2nd edition, Springer –Verlag Berlin, New York, **1996,** 212.
[16] RP Gujjeti, E Mamidala, *International Journal of Innovative Research in Science, Engineering and Technology*, **2013,** 2(10): 5725-5730.