

PHYTOCHEMICAL SCREENING AND THIN-LAYER CHROMATOGRAPHIC STUDIES OF *LITSEA GLUTINOSA* (LOUR.) BARK EXTRACT

Bharti Jain*¹, Ashita Rawat¹, Asna Mariyam¹, Geeta Parkhe²

1. Department of Chemistry, Sarojini Naidu Government Girls P.G. (Autonomous) College, Bhopal (M.P.)
2. Scan Research Laboratories, Bhopal (M.P.)

bhartijain@yahoo.com

Abstract

The present study deals with the phytochemical screening and thin-layer chromatographic studies of *Litsea glutinosa* bark extract belonging to family Lauraceae. Phytochemical screening determination by some chemical tests and thin layer chromatographic study was carried out by using various solvent system of varying polarity. Phytochemical screening reflects the presence of alkaloids, glycosides, flavonoids, diterpenes, phenols, amino acids, carbohydrate, proteins and saponins hydroalcoholic extracts. Thin layer chromatographic studies of the *Litsea glutinosa* bark extracts constituted different colored phytochemical compounds with different R_f values. The result obtained in present study indicated *Litsea glutinosa* bark as a rich source of natural antioxidants and provides evidence that solvent extract of *Litsea glutinosa* bark contains medicinally important bioactive compounds and this justifies the use of plant species as traditional medicine for treatment of various diseases.

Keywords: *Litsea glutinosa*, Bark extract, Phytochemical, Lauraceae, Retention factor, TLC.

INTRODUCTION

India has one of the oldest, richest and most diverse cultural traditions associated with the use of medicinal plants. Medicinal plants are great importance to the health of individuals and communities in general. The medicinal value of plants lies in some chemical substances that produce a definite physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids and phenolic compounds. Many of the indigenous medicinal plants are used as spices and food plants. They also sometimes added to foods meant for pregnant women and nursing mothers for medicinal purposes.¹⁻³ Herbs being easily available to human beings have been explored to the maximum for their medicinal properties.

Litsea glutinosa (Lour.) C.B. Rob is an aromatic tree belongs to the family Lauraceae and is found to be sparsely distributed in the Western Ghats, India. *L. glutinosa* is an evergreen medium-sized tree and plant can attain a height of 20 meters.⁴ *L. glutinosa* contain phytoconstituents like alkaloids, glycosides, flavonoids, saponins, tannins, phenolic compounds etc. The bark of *L. glutinosa*, “is one of the most popular of native drugs”, is considered to be capable of relieving pain, arousing sexual power and good for the stomach in the treatment of diarrhea and dysentery. *L. glutinosa* is widely used as a demulcent and as an emollient. The phytochemical constituents of the bark of *L. glutinosa* have been shown to possess effective antibacterial and antifungal activity.⁵ This species is critically endangered.⁶ Sap of fresh bark or

its decoction is prescribed as a remedy for diarrhea, dysentery, rheumatism, and as an aid to longevity. In addition, in current usage, a paste prepared by grinding bark with water is used as a plaster in cases of sprain, bruises, wounds, inflammation, back pain, rheumatic and gouty joints, bone fractures etc. It has analgesic, antiseptic and emollient effects.⁷ To identify the bioactive compounds responsible for the above pharmacological activities, phytochemical studies have been carried out and *L. glutinosa* bark was selected on the basis of its traditional medicinal.

MATERIALS AND METHODS

Collection of plant

Litsea glutinosa plant barks were collected from the rural areas of Bhopal (M.P), India, in the month of Oct - Nov., 2016.

Preparation of plant extract

Barks were collected in the shade dried and coarsely powdered after drying. The powdered material was subjected to extraction with petroleum ether by Soxhlet extraction procedure. The extraction was continued till the defatting of the material had taken place. After defatting, the plant material was subjected to hydroalcoholic extraction. The extract was filtered and it was finally dried at low room temperature under pressure in a rotary vacuum evaporator (Thermotech, Buchi type model th-012). The extracts were concentrated, percentage yield calculated and then subjected to phytochemical screening and TLC profiling studies. The dried extract was properly stored in the desiccators for further experiment and analysis.⁸

Phytochemical Screening

Chemical tests for the screening and identification of bioactive chemical constituents like alkaloids, carbohydrates, glycosides, saponins, phenolic compounds, phytosterols, proteins, amino acids, flavonoids, and tannins, in the medicinal plants under study were carried out in extracts by using standard procedure in.^{9,10}

Thin layer chromatographic studies

Each solvent extract was subjected to thin layer chromatography (TLC) as per conventional one dimensional ascending method using silica gel 60F254, 7X6 cm (Merck) were cut with ordinary household scissors. Plate markings were made with soft pencil. Glass capillaries were used to spot the sample for TLC applied sample volume 1-micro litre by using capillary at distance of 1 cm at 5 tracks. In the twin trough chamber with different solvent system Hexane: Acetic acid (9:1) solvent system I, In solvent system II Hexane: Ethyl acetate :Acetic acid (5:4:1), In solvent system III Hexane: Ethyl acetate: Acetic acid (4:4:2), In solvent system IV Hexane: Ethyl acetate: Acetic acid (3:6:1), In solvent system V Hexane: Ethyl acetate: Acetic acid (2:7:1) used. After pre-saturation with mobile phase for 20 min for

development were used. After the run plates are dried and sprayed freshly prepared iodine reagents were used to detect the bands on the TLC plates. The movement of the active compound was expressed by its retention factor (R_f), values were calculated for different samples

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

RESULTS AND DISCUSSION

The crude extracts so obtained after the extraction process was concentrated to obtain the actual yield of extraction. The yield of extracts obtained using different solvents are depicted in table 1.

Table 1: Result of percentage yield of different extract

S. No.	Solvents	Percentage Yield (%)
1.	Pet. Ether	1.2%
2.	Hydro-alcoholic	1.4%

A small portion of the dried extracts was subjected to the phytochemical test using methods to test for alkaloids, glycosides, tannins, saponins, flavonoids and steroids separately for extracts of all samples. The outcomes of the results of extract subjected to the phytochemical test are discussed in table 2.

Table 2: Result of phytochemical screening of extract

S. No.	Constituents	Hydroalcoholic Extract
1.	Alkaloids	+
2.	Glycosides	+
3.	Flavonoids	+
4.	Diterpenes	+
5.	Phenolics	+
6.	Amino Acids	+
7.	Carbohydrate	+
8.	Proteins	+
9.	Saponins	+

+ indicates presence

The present study carried out in the *Litsea glutinosa* revealed the presence of active medicinal constituents. The active phytochemical compounds of *Litsea glutinosa* were qualitatively analyzed for bark and the results are presented in Table 2. Among these phytochemical screening, alkaloids, glycosides,

flavonoids, diterpenes, phenols, amino acids, carbohydrate, proteins and saponins were present in hydroalcoholic extracts.

Table 3: Calculation of R_f Value

S. No.	Compound	Extract	R _f Value (Std.)
1.	Gallic acid	Toluene: Ethyl acetate: Formic acid (7:5:1)	0.65
2.	Quercetin	Toluene: Ethyl acetate: Formic acid (5:4:1)	0.82

A large number of solvent systems were tried to achieve a good resolution. Finally, the solvents toluene: ethyl acetate: formic acid {(7:5:1) and (5:4:1)} was used for thin layer chromatographic studies of the hydroalcoholic extract of *Litsea glutinosa*. Solvent system I toluene: ethyl acetate: formic acid (7:5:1), spots were visible with R_f values 0.65 for extract as well as standard gallic acid and solvent system II toluene: ethyl acetate: formic acid (5:4:1), 1 spot detected with R_f value 0.82 for extract and standard quercetin.

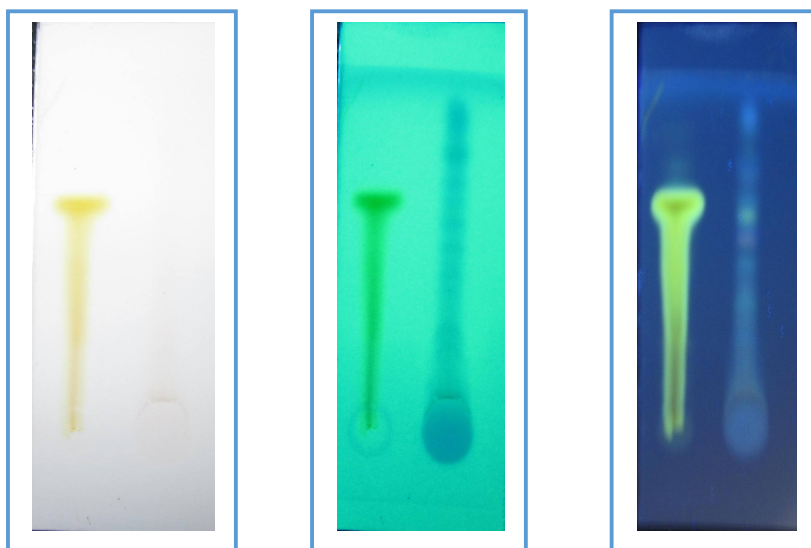


Figure 1: Thin layer chromatography of extract and standard (Gallic acid)

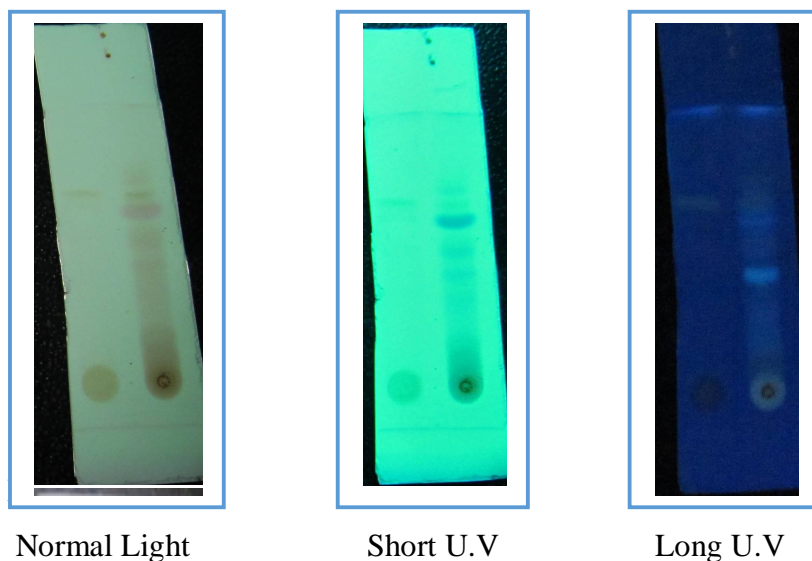


Figure 2: Thin layer chromatography of extract and standard (Quercetin)

A large number of plants produce secondary metabolites such as alkaloids, flavanoids, phenols, terpenoids and steroids that are used in pharmaceuticals, cosmetics and pesticide industries. In the present study, phytochemical screening of extract showed significant indication about the presence of metabolites. The results of the present study also supplement the folkloric usage of the studied plants which possess several known and unknown bioactive compounds with bio-activity. By isolating and identifying these bioactive compounds new drugs can be formulated to treat various diseases and disorders.

TLC profiling of extracts gives an impressive result that directing towards the presence of a number of phytochemicals. This information will help in the selection of an appropriate solvent system for further separation of the compound from these plant extracts.

Conclusion

The plant screened for phytochemical constituents seemed to have the potential to act as a source of useful drugs and also to improve the health status of the consumers as a result of the presence of various compounds that are vital for good health. The bark of *Litsea glutinosacana* provide lead molecules which could be a useful substrate for the synthesis of new broad-spectrum antibiotics for the treatment of infections caused by the organisms. Further purification, identification and characterization of the active compounds would be our priority in future studies.

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