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RESEARCH ARTICLE

PHYTOCHEMICAL SCREENING AND THIN-LAYER CHROMATOGRAPHIC STUDIES OF *GLORIOSA SUPERBA* LINN. SEED EXTRACT

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ABSTRACT

Gloriosa superba seed Linn. is one of the important medicinal plants now in endangered list. This plant is widely used for several ethano-medicinal purposes by tribal peoples and traditional practitioners. The present study deals with the phytochemical screening and thin-layer chromatographic studies of *Gloriosa superba* seed extract belonging to family Colchicaceae. 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, flavonoids, diterpenes and proteins in hydroalcoholic extract. Thin layer chromatographic studies of the *Gloriosa superba* seed extracts constituted different colored phytochemical compounds with different Rf values. The result obtained in present study indicated *Gloriosa superba* seed as a rich source of natural antioxidants and provides evidence that solvent extract of *Gloriosa superba* seed contains medicinally important bioactive compounds and this justifies the use of plant species as traditional medicine for treatment of various diseases.

Keywords: *Gloriosa superba*, Seed extract, Phytochemical, TLC.

INTRODUCTION

Herbs being easily available to human beings have been explored to the maximum for their medicinal properties. *Gloriosa superba* is a species of flowering plant in the family Colchicaceae. English language common names include flame lily, climbing lily, creeping lily, glory lily, gloriosa lily, tiger claw and fire lily. *Gloriosasuperba L.* is not only a notorious human and livestock poison, but is also widely used in several indigenous systems of medicine for the treatment of various human ailments. *G. superba* has caused illnesses and even fatalities to humans and animals due to both intentional and accidental poisoning. It is a native to tropical Africa, India and south-eastern Asia,[1] now widely cultivated throughout the world as an ornamental plant. It is common in forest-savanna boundaries, locally common in thickets, hedges, open forest, grassland and bush land, where it can be seen scrambling through other shrubs.[2] The generic name *Gloriosa* means 'full of glory' and the specific epithet *superba* means 'superb', alluding to the striking red and yellow flowers. All parts of the plant, but especially the tubers (swollen, underground stems), are extremely poisonous and the ingestion of flame lily has caused many accidental deaths. It has also been used to commit murder, suicide, to induce abortions and to poison dogs. African porcupines and some moles are reputed to be able to consume the roots with no ill effects.

To identify the bioactive compounds responsible for the above pharmacological activities, phytochemical and chromatographic studies have been carried out and *Gloriosa superba* seed was selectedon the basis of its traditional medicinal.

Materials and methods

Collection of plant

Gloriosa superba seeds were collected from the rural areas of Bhopal (M.P), India, in the month of December 2016.

Preparation of plant extract

Gloriosa superba seed were collected, 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 deffating, 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.

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. [3, 4]

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 (Rf), values were calculated for different samples. [5]

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

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.	Hydro-alcoholic	1.6%

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

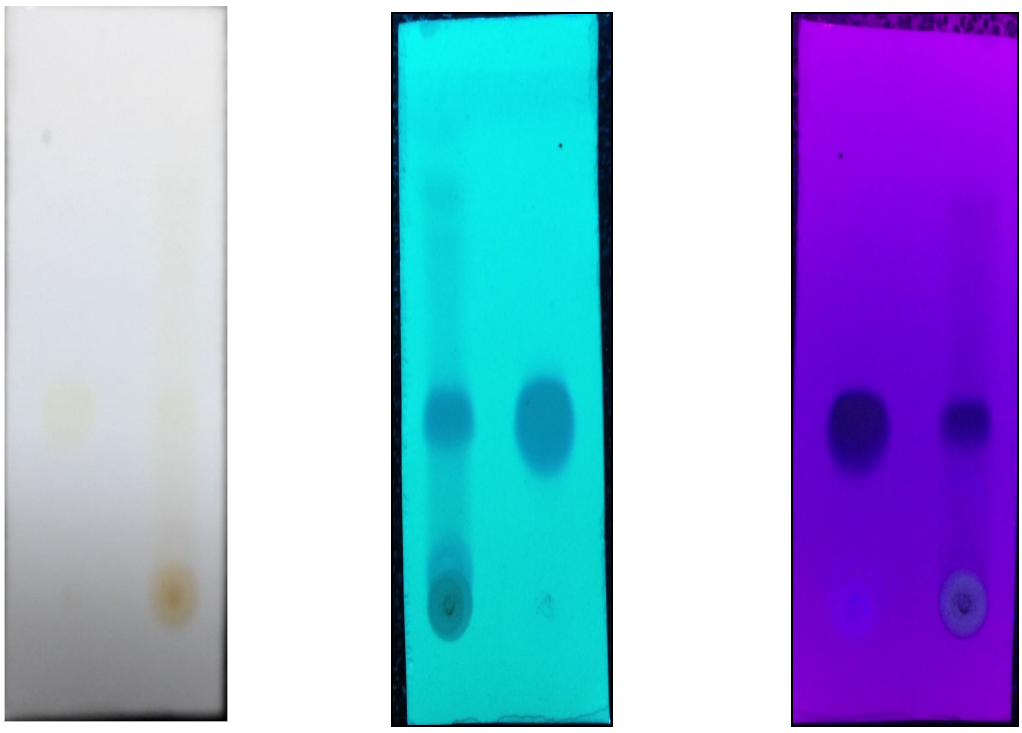
S.No	Phytochemical	Test	Results
1	Alkaloids	Wagner's test	positive
		Hager's test	positive
		Dragendroff's test	positive
2	Carbohydrates	Fehling test	negative
3	Saponin	Lather formation	negative
4	Phenols	Ferric chloride test	negative
5	Flavonoids	Lead acetate test	positive
6	Protein	Xanthoproteic test	positive
7	Diterpenes	Copper acetate test	positive

The present study carried out in the *Gloriosasuperba*revealed the presence of active medicinal constituents. The active phytochemical compounds of *Gloriosasuperba*were qualitatively analyzed for bark and the results are presented in Table 2. Among these phytochemical screening, alkaloids, flavonoids, diterpenes and proteins were present in hydroalcoholic extract.

From the Rf value it was confirmed the presence of colchicines as alkaloid compound in the extract

Table 3: Calculation of R_f Value

S. No.	Compound	Extract	Rf Value (Std.)
1.	Alkaloid	Ethyl acetate: methanol (10:1.3)	0.65



Normal Light **Short U.V** **Long U.V**

Figure 1: Photograph of TLC (Alkaloid)

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 *Gloriosasuperba* seed can 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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