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RESEARCH ARTICLE

BIOACTIVE COMPOUNDS ANALYSIS TUBER AND SEED OF Gloriosa *superba* GC –MS METHOD

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ABSTRACT

The present study deals with the phytochemical and GC- MS analysis of the medicinal plant of *Gloriosa superba*. The methanol extract of this plant showed good phytochemicals and bioactive compounds found in GC – MS analysis of *Gloriosa superba*. The result showed that 5 bioactive compounds in tuber and 4 bioactive compounds in seed are present. The alkaloid content of Colchicine highly present in seed

INTRODUCTION

Gloriosa *superba* is considered as an important medicinal plant and very much used in Indian system of medicine. *Gloriosa* popularly known as *"Glory lily"*. The family Liliaceae. *Glory lily* a perennial tuberous climbing herb is widely distributed in tropical and sub tropical parts of India including foothills of Himalayas (kapoor, 2001). It is known by different names in India, such as kalihari, Agnishikha, Languliata and Nangulika. It is being locally called as *"kanvali kizhangu"*, *"karthigai kizhangu* or *kalappai kizhangu"* in tamil. Germination of *Gloriosa* seeds by September to October and flowering is noticed from November to December (swarnapriya *et al.,* 1995). This herb is a native of tropical Asia and Africa and found growing throughout tropical India upto an altitude of 2500m (chopra *et al.,* 1956). *Glory lily* is a large glabrous, herbaceous branching climber with narrow leaves ending in spirally twisted climbing leaf tip tendrils. It arises from a perennial, fleshy tuberous rhizome. *Gloriosa superba* is a tuberous plant with V or L shaped, finger-like tubers that are pure white when young, becoming brown with age. It is one of the most important medicinal plants of Asia and Africa (sivakumar *et al.,* 2000, Jana *et al.,* 2011). Almost all parts of it find diverse medicinal usage (kapoor, 2001). It has been a well known plant in Indian Ayurveda and pharmacological industries as well (Asolkar *et al.,* 1992). It is mainly due to the presence of alkaloids like Colchicine (C_{22} H_{25} O_6 N) and its derivative like Gloriosin and Colchicocide (C_{27} H_{33} O_{11} N) along with Benzon acid, Salicylic acid, Sterols and resinous substances and therefore, the demand of this plant is increasing day by day. It is used in ayurvedic medicine as abortifacient, anti gout, anti leprotic, antipyretic, thermogenic and also anti cancerous agent. It is also provides relief to the swollen joints and in gout. In the Indian system of medicine, the tubers are used as tonic, antiperiodic, anthelmintic and also for snake bites (Gupta *et al.,* 2005). *Gloriosa* was only found in the wild a decade back but now it has been domesticated for economic gain and all parts of the plant are utilized in Indian medicine. The root is used as a germicide, to cure ulcers, piles, hemorrhoids, inflammation, scrofula, leprosy, dyspepsia, worm's infestation, flatulence, intermittent fevers, debility arthritis and against snake poison.

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MATERIALS AND METHODS

Collection of plant

The fresh tuber and seed of *Gloriosa superba* were collected in the fields of Jayamkondam area, Ariyalur District, Tamil Nadu. The tuber were cleaned of adhering soil/dust in the field by shaking and quick rinsing with tap water. Any remaining particles of soil were removed by use of pressurized air flow and by the use of paint brush and in some cases, by quick rinsing with distilled water. Tuber and seed were placed in paper bag and transferred to the laboratory.

Extraction

About 5grams of dried tuber and seed powder were extracted with 50 ml of methanol for 24 h at room temperature by constant shaking and filtered twice through Whatman no. 1 filter paper with the aid of a suction pump. Then the solvents were evaporated in water bath at 40ºC and the residue were transferred to screw cap bottles and stored in refrigerator until use.

Qualitative evaluation of phytochemicals

The preliminary screening test were performed for the presence of following secondary metabolites such as alkaloid, glycosides, terpenoids, tannin, flavonoids, saponins, steroid and phenols (Harborne, 1973).

Alkaloids test

The plant extract was evaporated to dryness and the residue was heated on a boiling water bath with 2% hydrochloric acid. After cooling, the mixture was filtered and treated with a few drops of Mayer's reagent. Formation of turbidity or yellow precipitation showed the presence of alkaloid.

Glycosides

Glycosides are compounds which upon hydrolysis give rise to one or more sugars (glycones) and a compound which is not a sugar (Glycone or Genine). To the solution of the extract in glacial acetic acid, few drops of ferric chloride and concentrated sulphuric acid are added, and observed for a reddish brown coloration at the junction of two layers and the bluish green colour in the upper layer.

Terpenoids and steroids

Four milligrams of extract was treated with 0.5 ml of acetic anhydride and 0.5 ml of chloroform. Then concentrated solution of sulphuric acid was added slowly and red violet colour was observed for terpenoid and green bluish colour for steroids.

Flavonoids

Four milliliters of extract solution was treated with 1.5 ml of 50% methanol solution. The solution was warmed and metal magnesium was added. To this solution, 5 – 6 drops of concentrated hydrochloric acid was added and red color was observed for flavonoids and orange colour for flavones.

Saponins

0.5 g of extracts was added to 5 ml of distilled water in a test tube. The solution was shaken vigorously and observed for a stable persistent froth. The frothing was mixed with 3 drops of olive oil and shaken vigorously after which it was observed for the formation of an emulsion.

Phenols

The extract (50 mg) is dissolved in 5 ml of distilled water. To this few drops of neutral 5% ferric chloride solution are added. A dark green colour indicates the presence of phenolic compounds.

Tannins

To 0.5 ml of extract solution 1 ml of water and 1-2 drops of ferric chloride solution was added. Blue colour was observed for gallic tannins and green black for catecholic tannins.

ESTIMATION OF PHOTOCHEMICAL ANALYSIS

The analysis of GC-MS technique is used to analyse the plant material. This important analytical technique is composed of gas chromatography (GC) and mass spectrometry (MS) which have been combined. The analysis of GC-MS identification different compounds available at Indian institute of technology, madras (IIT).

GC-MS Condition

GC-MS was performed with Hewlett-Packed Compounds were separated on a 30m x0.25mm capillary column coated with a 0.25 µM film of HP-5- sample were injected with a split ratio of 50:1; helium was used as carrier gas at 1.0 ml min^{-1}. The column temperature was maintained at 100°c for 1 minutes, after injection then increased at 10° min^{-1} to 275°c which was sustained for 20 minutes. The time required for chromatography of one sample 40 minutes.

Analysis of the phytocomponents in Gloriosa superba tuber and seed using GC-MS technique

One micro litre of the filtrate was injected into the GC-column. There the sample get evaporated and carried away by the carrier gas, helium and it get segregated into individual components. The sample fraction coming out of the column was let into the mass detector and the mass spectrum of each component was recorded. The mass spectrum of the unknown component was compared with the known spectrum was accomplished using computer searches in commercial libraries.

RESULT AND DISCUSSION

The study was designed to evaluate the phytochemicals and GC-MS analysis tuber and seed of *Gloriosa superba.* Methanol extract of *Gloriosa superba* showed good result for phytochemicals. The tuber containing saponins and glycosides are negative result other phytochemicals are positive result (Table 1).

Table 1 Pytochemical analysis in tuber of *Gloriosa superb*

SI. NO	Parameters	Result
1.	Flavonoids	+
2.	Alkaloids	+
3.	Tannins	+
4.	Phenol	+
5.	Steroids	+
6.	Saponins	-
7.	Glycosides	-
8.	Terpenoids	+

The seed good Phytochemicals like phenols, Alkaloids, tannins, flavonoids, terpenoids, steroids, saponins (Table 2).

Table 2 Pytochemical analysis in seeds of *Gloriosa superba*

SI. NO	Parameters	Result
1.	Flavonoids	+
2.	Alkaloids	+
3.	Tannins	+
4.	Phenol	+
5.	Steroids	+
6.	Saponins	+
7.	Glycosides	-
8.	Terpenoids	+

Table 3 Effect of rhizobacterial inoculation on Bioactive compounds present in tuber of *Gloriosa superba* by GC-MS

SI. NO.	RT	Compound Name	Molecular formula	Molecular weight
1.	11.46	Pyrrole-2-carboxylic acid,4-(1-chlorodec-1-enyl)-3,5-dimethyl,ethyl ester	$C_{19}H_{30}CINO_2$	212.3
2.	14.76	1,9-Dioxa-5-thianonane,3,7-bis(9-borabicyclo(3.3.1)non-9-yloxy)-1,9-diphenyl	$C_{34}H_{48}B_2O_4S$	574.42
3.	14.92	Hexadecanoic acid, 14 – methyl, methyl ester	$C_{18}H_{36}O_2$	284.47
4.	15.29	Colchicine	$C_{22}H_{25}O_6N$	399.44
5.	15.75	Ethanoeperoxoic acid,1-cyano-1-(2-(2-phenyl-1,3-dioxolan-2-yl)ethyl)pentyl ester	$C_{19}H_{25}NO_5$	347.40

Table 4 Effect of rhizobacterial inoculation on Bioactive compounds present in seed of *Gloriosa superba* by GC-MS

SI. NO.	RT	Compound Name	Molecular formula	Molecular weight
1.	7.82	Pentadecanoic acid, 14-methyl, methyl ester	C_{17} H_{34} O_2	270.45
2.	8.02	(2-((4-methoxyphenyl)imino)-4-methy-1,3-thiazolan-4-yl)methanol	C_{12} H_{16} N_2 O_2 S	252.33
3.	8.40	Hexadecanoic acid , ethyl ester	C_{18} H_3 $6O_2$	284.00
4.	8.73	Colchicine	C_{22} H_{25} O_6 N	399.44

The GC – MS analysis in tuber and seed of *Gloriosa superba* . The tuber contains 5 major peaks. In that peak of bioactive components were identified and tabulated (Table 3 and figure 1). The seed containing 4 major peaks. In that peak of bioactive components were identified and tabulated (Table 4 and figure 2) with compound name, molecular formula and molecular weight. The major compound of colchicine present in tuber and seed of *Gloriosa superba* plant.

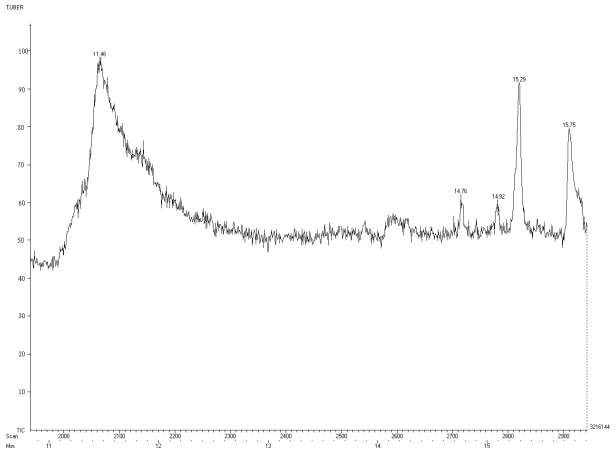

Figure 1 Effect of rhizobacterial inoculation on Bioactive compounds present in tuber of *Gloriosa superba* by GC-MS

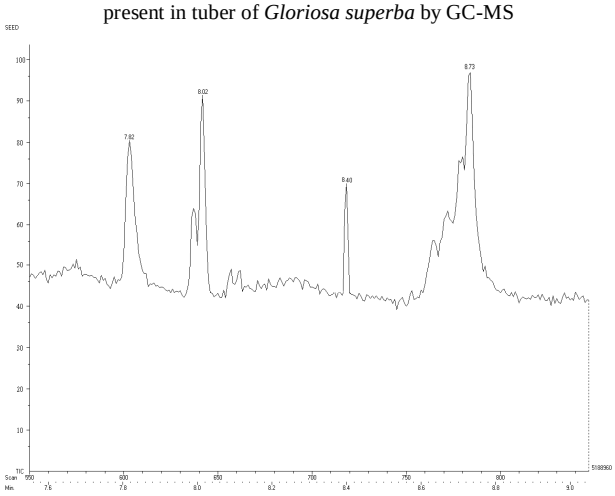

Figure 2 Effect of rhizobacterial inoculation on Bioactive compounds present in seed of *Gloriosa superba* by GC-MS

CONCULSION

Plants are natural source of bioactive compounds to treat many diseases. The plant *Gloriosa superba* has showed good phytochemicals which means that it can use for treating diseases. In GC-MS analysis 5 bioactive compounds for tuber and 4 bioactive compounds for seed were identified. The main alkaloid content of colchicine used for treating many diseases. The result may have bioactive compounds prevention of skin related diseases.

Reference

Asolkar, L.V., K.K. Kakkar, O.J. Chakare. 1992. Second supplement to Glossary of Indian Medicinal plant with Active principles. Part-I (A-K). Publications and Information Directorate, council of scientific and Industrial Research, New Delhi

Chopra, R.N., S.L. Nayar and I.C. Chopra. 1956. Glossary of Indian medicinal plants. pp. 125-126.

Gupta, L.M., RC. Rana, R. Raina, M. Gupta. 2005. Colchicines content in *Gloriosa superba L.* Jour. Rese. (SKUAST- Journal), 4: 238 -241.

Jana, S., G.S. Shekhawat. 2011. Critical review on medicinally potent plant species : *Gloriosa superba*. Fitoterapia., 82 (3) : 293 – 301.

Kapoor, L.D. 2001. Traditional uses of medicinal plant In: Ayurvedic medicinal plant. CRC press, New Delhi.

Sivakumar,G., and K.V. Krishnamurthy. 2000. Micropropagation of *Gloriosa superba L.* an endangered species of Asia and Africa. Curr. Sci., 78: 1-10.

Swarna priya, R., A. Doraipandian, T. Arumugam and N.S. Radha. 1995. Floral biology of *Gloriosa superba L.* south Ind. Horti., 43 (1-2)**:** 40 - 41.

Arbone JB. 1973. Phytocemical methods, London.chapman and Hall, ltd. 49 – 188.

Alok P Jain, Satish Suriyavanshi. 2010. *Gloriosa superba linn.* A pharmacological Review. Int. Jour . Pharmaceuti. Rese. Develop., 2(8), 004: 24 – 29.

Ade, R., M.K. Rai. 2009. Review : current advances in *Gloriosa superba* L. Biodivers., 10 (4) : 210 – 214.

Amerakpam Nirjanta Devi and W. Femina. 2012. GC-MS analysis of *Gloriosa superba* medicinal plant of Tamilnadu. Int. Jour. Pharma. Rese., 5(1) : 343 – 345.

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