

Qualitative and quantitative analysis of *Gloriosa Superba* L

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Abstract

Gloriosa superba L. belonging to family *Colchicaceae* is very unique plant having large number of medicinal values. It is a perennial herb growing from a seed or fleshy rhizome. Present study on qualitative and quantitative analysis of *Gloriosa superba* L. The qualitative phytochemical studies were carried out in the solvents viz. Methanol, Chloroform and n-Butanol. The Methanol, Chloroform and n-Butanol tuber and leaves extract of *Gloriosa superba* L. shows the presence of alkaloid, glycosides, terpenoids, tannin, flavonoids, saponins, steroid and phenols but in tuber of *Gloriosa superba* L. high intensity of phytochemical than that of leaves. also, the quantitative studies were carried out in the same solvent mentioned above, Alkaloid content in tuber of *Gloriosa superba* L. that was 2.921, 2.546 and 3.045 $\mu\text{g/ml}$ respectively, and total flavonoids in tuber, 0.845, 0.641 and 0.978 $\mu\text{g/ml}$ respectively and also followed by total content of phenols that was 1.284, 0.652 and 1.361 $\mu\text{g/ml}$.

Keywords: qualitative, quantitative, *Gloriosa superba* L., n-Butanol etc

1. Introduction

Natural products or phytochemical are still recognized, as one of the most important resource of bioactive compounds. Currently, more than half of the world's population relies on plants as the unique source of remedies, with which to treat a wide variety of disorders. Additionally, around 40-80% of new drugs, approved and under commercialization are derived from natural products. The four major classes of secondary metabolites are Terpenoids, Phenols, Glycosides, and Alkaloids. Phytochemical technique mainly applied to the quality medicines of various chemical components such as alkaloids, flavonoids, saponins, phenolics, terpenoids, tannins, etc. Phytochemicals from medicinal plants are receiving even greater attention in the medical science. In the developing countries, over the counter remedies and "Ethical Phytomedicines", which are standardized toxicologically and clinically define crude drugs, are seen as a promising low-cost alternative in primary health care. The field also has benefited in recent years from the interaction of the study of traditional ethnobotanical knowledge and the application of modern phytochemical analysis and biological activity studies to medicinal plants.

So, I have selected present study on qualitative and quantitative analysis of *Gloriosa superba* L. is an important medicinal plant belonging to *Colchicaceae* family. It is a semi-woody herbaceous branched climber reaching plant approximately 5 meters height, with brilliant wavy-edged yellow and red flowers. It is one of the endangered species among the medicinal plants and Different parts of the plant have a wide variety of uses especially within traditional medicine practised in tropical Africa and Asia. The tuber is used traditionally for the treatment of bruises and sprains, colic, chronic ulcers, haemorrhoids, cancer, impotence, nocturnal seminal emissions, leprosy also for inducing labour pains and abortion. Haroon, *et al.*, 2011, the tuberous root of *G. superba* boiled with sesamum oil is applied twice a day on the joints, affected with arthritis reduces pain. It is also used in wounds, skin related problems, fever, piles,

inflammation, uterine contractions, blood disorders, general body toner and poisoning. Some researcher reported pharmacological properties of *Gloriosa superba* L. Hemaiswarya, *et al.*, 2009 reported that Tubers extract in Methanolic, aqueous and petroleum ether shows Antibacterial, antifungal, and mutagenic activities, and Kumarappan, *et al.*, 2011^[3] reported that tubers extract in Alcohol shows Antihemolytic activities.

2. Material and Methods

2.1 Collection of Plant parts

Collections of *Gloriosa superba* L. plant part were collected from Gogababa Tekkadi, Dr Babasaheb Ambedkar Marathwada university campus Aurangabad Maharashtra state, India. Plant part tuber and leaves cleaned soil dust with tap water then dried under shade and prepared fine powder and kept it in airtight bottle. The plant materials were identified by using standard floras like Cook 1907, Dhore 2005, Naik 1989, Yadav and Sardesai 2002.

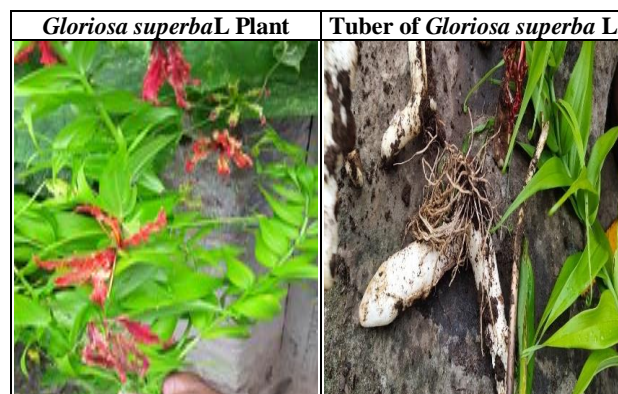


Fig 1

2.2 Preparation of Plant Part Extract

Methanol, Chloroform and n-Butanol, extract was prepared by using Soxhlet extractor. 30 gm of each plant part powder

was placed in a thimble, which was placed in chamber of the Soxhlet apparatus. 300 ml solvent in the flask and the temperature was maintained at 55 °C for 72 hours. Then the extracts were filtered through Whatman filters paper No 1. Solvent was evaporated at 40-50 °C by using Rotary evaporator. The collected powder was weight and dissolved in Dimethyl sulfoxide (DMSO) with 10% concentration. The extracts were used for Qualitative and quantitative evaluation of phytochemical. (Handa *et al.*, 2008, Subramanian *et al.*, 2011) ^[9, 8].

2.3 Qualitative analysis of *Gloriosa superba* L. plant parts

The qualitative screening test were performed for the presence of following secondary metabolites such as alkaloid, glycosides, terpenoids, tannin, flavonoids, saponins, steroid and phenols (Harborne, 1973) ^[5] and Sofowara (2005).

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 were 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

4 ml 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 colour 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. 3

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.

2.4 Quantitative analysis of *Gloriosa superba* L. plant parts

Preparation of plant extracts for quantitative determination of alkaloids 5 gm of powdered plant material was taken into 20 ml of n-butanol and vigorously stirred. The content was transferred into a reagent bottle. The slurry was kept overnight at room temperature. Then it was centrifuged at 6000 rpm for 10 min and the supernatant was made up to 50 ml with n-butanol

2.4.1 Estimation of total alkaloids by titrimetric methods used by Plummer, 2013 ^[4] and Debnath *et al.* 2015 ^[2].

Obtained supernatant of the plant sample was used for the estimation of total alkaloids by titrimetric methods. 10 ml of the supernatant was taken into a 100 ml separating funnel. 10 ml of 0.1 (N) HCl was added and shaken thoroughly for 2-3 min. This results in the solubility of alkaloids. The lower layer contains alkaloids neutralized with 0.1 (N) HCl and the upper layer contains n-butanol. 10 ml HCL portion was collected in a beaker and 2-3 drops methyl red was added to it, that turns the solution into slightly reddish colour. The contents of beaker were titrated against 0.1 (N) NaOH, till colour change changed from red to pale yellow. The neutralization point was determined. Same procedure was repeated triplicate. The total amount of alkaloids was calculated by considering the following equivalent:

1 ml 0.1N HCl \equiv 0.0162 g alkaloid

2.4.2 Estimation of Total Phenolic Content

Total phenol content of *G. Superba* L. was assayed by modified Dewanto *et al.*, 2002 ^[13] and Jothi *et al.*, 2019 ^[12] procedure. The different concentrations of 10 μ g, 20 μ g, 40 μ g, 60 μ g, 80 μ g, and 100 μ g were using an aliquot of diluted extract and added to 0.25 ml of Folin Ciocalteu reagent. The elucidation was adjusted with distilled water to a final volume of 3ml and shaken thoroughly. The solution was incubated and kept in the dark placed and read at 760 nm was read against prepared blank. The total phenol content of plant parts was expressed as milligrams of gallic acid equivalents per gram of dry weight. The total sample was analyzed in three replicates.

2.4.3 Estimation Total Flavonoid Content

Total Flavonoid content in *G. Superba* L. whole plant extract was analyzed by the aluminium chloride colorimetric system M.M Mervat, *et al* 2009 ^[7] and Jothi *et al.*, 2019 ^[12]. 0.5ml of plant part extract of at different concentrations like 10 μ g, 20 μ g, 40 μ g, 60 μ g, 80 μ g, and 100 μ g were taken and the final volume was made up to 3ml with methanol. After that, 0.1ml AlCl₃ (10%), 0.1ml of potassium acetate and 2.8ml of distilled water were added continuously and test solution was vigorously shaken. After 30 minutes for the incubation periods, absorbance was recorded at 415 nm. The concentration of flavonoids in test samples was calculated and expressed as the equivalent of quercetin (QE) / g of sample. The entire sample was analyzed in three replicates.

3. Results and Discussion

Table 1: Qualitative analysis of *Gloriosa superba* L. Tuber and leaves

Sr. No.	Phytochemical	Plant extract of tuber			Plant extract of Leaves		
		Methanol	Chloroform	n-Butanol	Methanol	Chloro-form	n-Butanol
1	Alkaloids	++	++	++	++	+	+
2	Glycosides	++	+	++	+	+	+
3	Terpenoids	+	++	+	+	+	+
4	Steroids	++	+	++	++	+	+
5	Flavonoids	++	++	++	++	+	+
6	Saponins	++	++	++	+	++	++
7	Phenols	++	+	++	++	+	-
8	Tannins	++	++	++	-	+	-

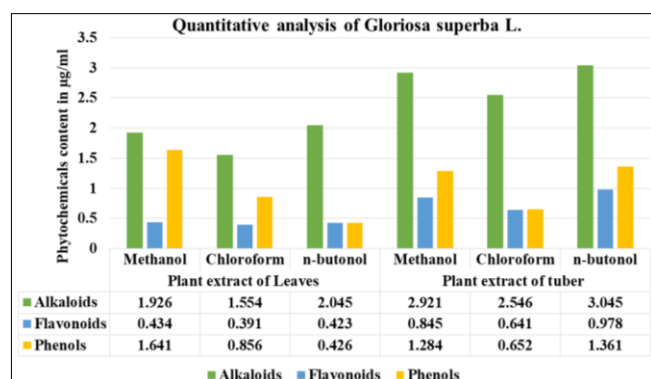


Fig 2: Quantitative analysis of *Gloriosa superba* L. plant parts, (leaves and tuber µg/ml)

Results taken average of triplicates for different concentration of plant extract.

The qualitative phytochemical studies were carried out in the solvents viz. Methanol, Chloroform and n-Butanol. The Methanol, Chloroform and n-Butanol tuber and leaves extract of *Gloriosa superba* L. shows the presence of alkaloid, glycosides, terpenoids, tannin, flavonoids, saponins, steroid and phenols but in tuber of *Gloriosa superba* L. high intensity of phytochemicals than that of leaves showed in table no.1 by twice and tannin absence in methanolic and n-butanol extract of leaves. Also, the quantitative studies were carried out in the same solvent mentioned above, Alkaloid content in tuber of *Gloriosa superba* L. that was 2.921, 2.546 and 3.045 µg/ml respectively, and total flavonoids in tuber, 0.845, 0.641 and 0.978 µg/ml respectively and also followed by total content of phenols that was 1.284, 0.652 and 1.361 µg/ml.

Total content of Alkaloid, flavonoids and phenols in the Methanol, 1.926, 0.434 and 1.641 µg/ml. respectively, in the Chloroform 1.554, 0.391 and 0.856 µg/ml. and followed by in the n-Butanol leaves extract of *Gloriosa superba* L. that was 2.045, 0.423 and 0.426 µg/ml.

4. Conclusion

Gloriosa superba L. is the rich source of phytochemicals, alkaloid, glycosides, terpenoids, tannin, flavonoids, saponins, steroid and phenols. Its extraction in n-butanol solvent shows highest intensity and content of phytochemicals followed by methanolic extract of tuber of *Gloriosa superba* L and it shows antimicrobial activities. Sagbo *et al*, 2005 and Jothi. 2019 [12], reported that Polyphenols and phenols found in plants are two secondary metabolites considered as natural antioxidants. Jothi, 2019 [12], and Trease *et al*, 1983 showed that *Gloriosa superba* is an alkaloid plant, which contains alkaloid components such

as colchicine and gloriosine which are mostly used in pharmaceutical formulation for drug. Noroozi *et al*, 1998 [6] and Al-Humaid *et al*, 2010 [1] reported Flavonoids are ketonic compounds that can induce anti-inflammatory activity and inhibits the oxygen compounds, enzyme cyclo-oxygenase dependent pro inflammation activity. Furthermore, flavonoids have a powerful anti-inflammation activity as they inhibit prostaglandin synthesis. Flavonoids in higher plants are inseparable with cardiovascular diseases and antioxidant potentials that can treat cancer disease. Pietta, 2000 and T. Sivakumar, 2017 [11] reported that Flavonoids and antioxidants origin of vitamins A, C, E and plant source diets.

So, *Gloriosa superba* L. of plant presence different phytochemical compounds useful for Further purification, identification and characterization of the active compounds of would be our priority in future studies.

5. References

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