



**PHYTOCHEMICAL EVALUATION AND FLUORESCENCE
ANALYSIS OF *SOLANUM NIGRUM* L.**

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ABSTRACT

Solanum nigrum L. belongs to the Solanaceae family has been used in traditional folk medicine. It has effects of antipyretic, diuretic, anticancer, anti-ulcer and hepatoprotective and therefore it has important medicinal values. **Objective:** The present study was aimed to investigate the preliminary phytochemical screening and fluorescence analysis of stem, leaf and unripened fruits of *Solanum nigrum* L. This study represents a report of qualitative determination of

phytochemicals of *Solanum nigrum* L. **Methods:** The phytochemical constituents like Alkaloids, Saponins, Tanins, Flavonoids, Carbohydrates and Proteins were investigated using Aqueous, Ethanol, Acetone and Chloroform extracts qualitatively. The fluorescence character of the plant sample was studied both in daylight and UV light after treatment with different reagents like methanol, petroleum ether, sodium hydroxide, hydrochloric acid, sulphuric acid, nitric acid and ferric chloride. **Results:** The qualitative phytochemical studies have clearly demonstrated that the plant *Solanum nigrum* L is a rich source of alkaloids, flavonoids, saponins, phenols, carbohydrates and protein. Aqueous extract of stem, leaves and unripened fruit contained more phytochemical constituents. The. Furthermore, Fluorescence analysis of stem, leaves and unripened fruits confirmed the standardization of quality, purity of the sample. **Conclusion:** Among the extracts, the aqueous extract has found to be rich in phyto constituents, which gave a strong reason to select this plant for pharmacological studies.

KEYWORDS: *Solanum nigrum* L., Phytochemical, Fluorescence, Standardization.

INTRODUCTION

Medicinal plants sources have been in usage in Homeopathy, Ayurvedic, Allopathy & Traditional medicine. Their use have been multiplied due to a number of side effects from the use of synthetic drugs, antibiotics and high cost. In developing countries 80% of the population still use traditional folk medicines obtained from natural resources.^[1] The curative properties of medicinal plants are mainly due to the presence of various complex chemical substance of different composition which occur as secondary metabolites.^[2] Phytochemical which posses many ecological and physiological roles are widely distributed as plant constituents, a great variety of phytochemicals includes alkaloids, flavonoids, tannins, phenolic compounds, saponins, lignins and lignans.^[3]

Solanum nigrum L. commonly known as Black nightshade is a dicot weed in the Solanaceae family. It is an annual herb, erect, 25 – 100cm high with a green, smooth and semi climbing stem. The opposite leaves with whole limb, oval and diamond shape are slightly cogged. Inflorescence are extra axillary umbels, the calyx cup – shaped, the corolla is white, 8- 10mm long, the lobes ovate – oblong pubescent abaxially, ciliate spreading. Filaments are 1 – 1.5 mm long, anthers oblong, 2.5 – 3.5mm, fruiting pedicels are strongly deflexed. The fruits are dull black, globose 8 – 10mm in diameter. It is a rather common species in near river, waste land, oil field and cultivated land. *Solanum nigrum* as a variety of pharmacological investigations including Anti-ulcer, anti-oxidant, Alzheimer's diseases and retinopathic diseases. The juice of the plant is used on ulcers & other skin diseases. The fruits are used as a tonic, laxative, appetite stimulant. The entire plant is used for anti – ulcerogenic effects.^[4]

Alkaloids content in *Solanum nigrum* is responsible for its medicinal value. Flavonoid in intestinal tract lowers the risk of heart diseases as they provide anti - inflammatory activity.^[3]

Saponin are a special class of glycosides which have soapy characteristics. It has a property of precipitating and coagulating red blood cells. some of the characteristics of saponin include formation of aqueous solution, cholestrol binding properties and bitterness.^{[7][3]}

Proteins are used for the building block of body tissues and promotes muscle growth.^[8]

Phenolic compounds exhibit a wide range of physiological properties, such as anti-allergenic, anti-athero-genic, anti-inflammatory, anti-microbial, antioxidant, anti-thrombotic, cardioprotective and vasodilatory effects^[9] Carbohydrates performs by providing energy and regulation of blood glucose level. Carbohydrates provide fuel to the central nervous system and muscular system. Carbohydrate diet that have low Glycemic index may improve insulin

sensitivity.^[10] Though many reports on stem, leaves are available in *Solanum nigrum*, the study of unripened fruits are negligible.

Many substances such as alkaloids like quinine and berberine in dilute sulphuric acid when suitably illuminated, emit light of different wavelength or colour from that which falls on them. This emitted light (fluorescence) ceases when the exciting light is removed.^[11] The fluorescence character of the plant powder was studied both in daylight and UV light after treatment with different reagents using methanol, ammonia, petroleum ether, sodium hydroxide, hydrochloric acid, sulphuric acid, nitric acid and ferric chloride. Hence the study was aimed to investigate the Phytochemical evaluation and fluorescence analysis of *Solanum nigrum* L. in stem, leaf and unripened fruit extracts.

MATERIALS AND METHODS

Collection of Sample

Solanum nigrum seeds were collected from an organic farm in Thiruvallur district. The seeds were sowed in pots and grown. 60days old fresh leaves, stems, and unripened fruits were collected separately and shade dried for 10 days and grounded using mortar and pestle. The dried powder of stem, leaves and unripened fruits was used for determination of phytochemical studies and fluorescence analysis.

Extraction of Plant Material

Preparation of extracts using Aqueous, Ethanol, Acetone and Chloroform

5 gm of dried sample (stem, leaves and unripened fruits) was weighed using electronic balance and was soaked in 25 ml of Aqueous, Ethanol, Acetone and Chloroform for 24hrs separately. The test tubes containing the extracts were boiled at 50 -60°C for 10mins and then filtered using Whatman No. 1 filter paper. The filtrate was then centrifuged at 2500rpm for 15minutes and the filtrate was collected in sterile bottles and stored in refrigerator at 5° C until further use.^[12]

Preliminary Phytochemical Analysis

Qualitative Phytochemical analysis of the crude powder of the *Solanum nigrum* L for the test of phytochemicals such as alkaloid, flavonoid, saponin, tannins and proteins and carbohydrates was carried out according to the methods described by^[12,13,14] is shown below.

Preliminary Phytochemical analysis**Alkaloids: (Mayer's test)**

To 1.0 ml of the extract few drops of Mayer's reagent was added. The observation of white precipitate indicates the presence of "Alkaloids"

Carbohydrates: (Molish test)

To 1.0 ml of extract few drops of Molish reagent was added. A brown ring (or) Violet ring appeared and gradually spread throughout the solution indicates the presence of Carbohydrates.

Flavonoids: (NaoH test)

To 1.0ml of extract few drops of 10% NaoH solution was added. Intense yellow colour was observed which turned colourless on addition of few drops of Conc.H₂SO₄. This indicates the presence of Flavonoids.

Phenols: (Ferric chloride test)

To 1.0 ml of extract few drops of ethyl alcohol and ferric chloride solution was added. Appearance of bluish green colour indicates the presence of Phenols.

Proteins: (Alkaline test)

To 1.0 ml of extract few drops of alkaline copper reagent & phenol reagent was added and kept in dark for few hours which turned to dark bluish green indicates the presence of Protein.

Quinines

To 1.0ml of extract few drops of NaOH was added. red coloration indicates the presence of Quinines.

Saponins: (foam test)

To 2.0 ml of extract 2ml of distilled water is added & shaken vigorously for the stable persistent froth indicates the presence of Saponin.

Tannins

To 1.0ml of extract 1.0ml 15% ferric chloride was added. A blue colour indicates Condensed tannins and green colour indicates Hydrosable tannins.

Glycosides

To 1.0ml of extract 3.0ml of chloroform and 10% ammonium solution was added.formation of pink colour indicates the presence of Glycosides.

Terpenoids: (salkowski test)

To 1.0 ml of extract 2.0 ml of chloroform, Conc.H₂SO₄ (3ml) was carefully added to form a layer. A reddish brown colouration of the interference indicated the positive results for the presence of Terpenoids.

Organoleptic evaluation

Organoleptic evaluation refers to the assessment of the selected plant by colour, odour, taste, texture. The organoleptic characters of the samples were evaluated according to the methods of.^[15]

Fluorescence analysis of plant material

The fluorescence character of the plant powder was studied both in daylight and UV light after treatment with different reagents like methanol, ammonia, Petroleum ether, sodium hydroxide, hydrochloric acid, sulphuric acid, nitric acid and ferric chloride. Fluorescence study is an essential parameter for the first line standardisation of crude drug. In Fluorescence the fluorescent light is always of greater wavelength than the exciting light. Light rich in short wave length is very active in producing fluorescence and this is the reason ultraviolet light produces fluorescence in many substances which do not fluorescence in daylight.^[16,17]

RESULTS

Table 1: Phytochemical analysis of Stem in *Solanum nigrum* L.

S.no	Parameters	Aqueous	Ethanol	Acetone	Chloroform
1	Alkaloids	+	++	--	--
2	Carbohydrates	++	++	++	++
3	Flavonoids	+	+	++	--
4	Phenols	++	+	--	++
5	Protein	++	--	--	++
6	Quinines	--	--	--	--
7	Saponins	++	++	+	+
8	Taninns	--	++	++	+
9	Glycosides	--	--	--	--
10	Terpenoids	--	--	--	--

++ = Abundantly present, + = present, -- = Absent

Table 1: Showed that the aqueous and ethanol stem extract of *Solanum nigrum* contains more variety of phytochemicals like Alkaloids, flavonoids, saponins, carbohydrates and proteins. Carbohydrates and saponin content are present abundant in all the extracts.

Table 2: Phytochemical analysis of leaves in *Solanum nigrum* L.

S.no	Parameters	Aqueous	Ethanol	Acetone	Chloroform
1	Alkaloids	++	--	++	++
2	Carbohydrates	++	++	++	--
3	Flavonoids	+	++	--	+
4	Phenols	++	++	++	++
5	Protein	++	+	--	--
6	Quinines	+	--	++	+
7	Saponins	++	+	+	++
8	Taninns	--	--	+	+
9	Glycosides	--	--	--	--
10	Terpenoids	++	--	+	+

++ = Abundantly present, + = present, -- = Absent

Table 2: Showed that the aqueous and acetone leaf extract of *Solanum nigrum* L contains an abundant variety of phytochemicals like Alkaloids, flavonoids, saponins, carbohydrates, proteins and terpenoids. Saponin and phenol content were present in all the extracts.

Table3: Phytochemical analysis of unripened fruit in *Solanum nigrum* L.

S.no	Parameters	Aqueous	Ethanol	Acetone	Chloroform
1	Alkaloids	++	--	++	++
2	Carbohydrates	++	--	--	++
3	Flavonoids	++	+	+	+
4	Phenols	++	--	++	+
5	Protein	++	++	+	--
6	Quinines	--	--	--	--
7	Saponins	++	++	++	+
8	Taninns	+	--	--	--
9	Glycosides	--	--	--	--
10	Terpenoids	--	+	--	--

++ = Abundantly present, + = present, -- = Absent

Table 3: Showed that the aqueous unripened fruit extract of *Solanum nigrum* L.contains a variety of phytochemicals like Alkaloids, flavonoids, saponins, carbohydrates, proteins and terpenoids. The results also reveals that the Saponins and Flavonoids are present in all the four extracts of *Solanum nigrum* L.

Organoleptic characters studies

The organoleptic characters of the *Solanum nigrum* L were determined and the results are shown in table 4.

Table 4: Organoleptic characters.

S.NO	Characters	Observation
	Stem	
1	Colour	Green
2	Odour	Characteristic
3	Taste	Bitter
	Leaf	
1	Colour	Dark green
2	Odour	Charaterstic
3	Taste	Slightly Bitter
	Unripened fruit	
1	Colour	Light green
2	Odour	Musk
3	Taste	Slightly Bitter

Fluorescence analysis

The results of fluorescence analysis were expressed in Table 5.

Table 5: Fluorescence Analysis of *Solanum nigrum* L.

S.No	Solvents Treatment	STEM		LEAF		FRUIT(UNRIPENED)	
		Visible	U V	Visible	UV	Visible	U V
1	Drug+distilledH ₂ O	Light green	Light green	Olive green	Dark Green	Light brown	Light green
2	Drug+Petroleum ether	Pinkish orange	Yellowish green	Yellowish green	Pinkish orange	Yellowish green	Pinkish red
3	Drug+chloroform	Yellowish green	Light green	Dark green	Reddish orange	Green	Reddish pink
4	Drug + Methanol	Dark green	Light green	Bright green	Pinkish orange	Light green	Ceramic yellow
5	Drug+conc .HCL	Blackish green	Dark green	Greenish black	Black	Brownish black	Brownish black
6	Drug+conc.HNO ₃	Ceramic yellow	Yellow	Yellowish green	Brown	Bright yellow	Brownish yellow
7	Drug+50%H ₂ SO ₄	Blackish brown	Black	Brownish black	Black	Brownish black	Brown
8	Drug + Ammonia solution	Light green	Yellowish Green	Dark green	Green	Yellowish green	Light green
9	Drug+10%NaOH	Light green	Ceramic yellow	Greenish black	Dark Green	Bright yellow	Light green
10	Drug + 5% FeCl ₂	Dark green	Bluish green	Greenish black	Blackish green	Dark green	Greenish black

DISCUSSION

The qualitative analysis of phytochemical constituents of stem, leaf and unripened fruit extract of *Solanum nigrum* L. reveals the presence of Alkaloid, Flavonoids, Saponin, Terpenoid, Carbohydrates and Proteins. The Aqueous extract of *Solanum nigrum* L contains more variety of phytochemicals in which the present study correlates with the previous study [F.O Atanu, U. G. Ebiloma 1 and E. I Ajay 2010]^[18] and it is Anti-ulcerogenic and Anti-inflammatory effects. Saponin content were present in all the extracts of *Solanum nigrum* L. Similarly the study of [A. Parvathi et al 2013]^[19] relates that the ethanolic extracts of solanaceae plants contain more amount of saponin content present in it.

The aqueous and ethanolic extract of the stem contains rich amount of Alkaloids content correlates to the previous study [Pronob Gogoi et al 2012]^[5] as an Anti-pasmodic and Anti-bacterial activities. Flavonoids, Phenols and Saponins constituents were present highly in all the leaf extracts. According to [A. Parvathi et al 2013]^[19] the phenols are found rich in ethanol, methanol and DMSO extracts of Solanaceae members.

The leaves shows wide range of phytochemical constituents present in it and the study is related to previous study [Pronob Gogoi et al 2012]. Flavonoids in intestinal tract lowers the heart diseases and as Anti-oxidants Flavonoids also provides Anti-inflammatory activity [D.E.Okwu et al 2004]. This may be the reason *Solanum nigrum* L have been used for treatment of burns and ulcers in herbal medicine. According to [Kumawat N.S et al 2010][Catopano -1997] [Kameswararao et al 1997]^[20] flavonoids are potent Anti oxidants and are known to modulate the activities of various enzymes due to the interaction with various biomolecules. It was reported that phytochemical of this plant possess bioactive anti-diabetic activities.

The aqueous extracts of unripened fruits contain more phytochemical constituents in it. The Saponin and Flavonoid content are found plenty in all the extracts of *Solanum nigrum*. L. This resembles that the study of [Akthar and munir, 1989]^[23] which reports that the ethanolic extracts of the unripened fruit of *Solanum nigrum* L was studied for its Neuropharmacological properties on experimental animal on intra peritoneal injection. The test extract also exhibited significant hydroxyl radical scavenging potential, thus by suggesting its probable mechanism of cytoprotection. Small unripened fruits of *Solanum nigrum* L. has a high concentration of solasodine but both the concentration and the absolute amount per fruit decreases with fruits maturation [Krithkar and Basu 1935, Nadkarni 1976].^[24] According to Cham B.E Wilson

1987].^[25] reports that unripened fruits contain Glycolakaloid which interfere with the membrane of the cells and they disrupt the integrity of cells upto a apoptosis especially in cancer cells.

The organoleptic studies of plant material helped for the confirmation and presence of some organic compounds due to the characteristic smell of the extracts and the bitter taste of fruit indicates that plant contains alkaloid compound in it. The fluorescence analysis of material under visible light and UV light was done. Some of the substance may be often converted into fluorescent derivatives by using different chemical reagents though they are not fluorescent, hence it may often assess qualitatively some crude drugs using fluorescence, as it is most important parameter.^[27]

Thus the results obtained in this investigation gives a clear indication of the presence of Alkaloids, Flavonoids, Saponins, Glycosides, Tannins, Carbohydrates and Proteins. Certainly the future investigation provides much light on beneficial properties, which could further economically exploit the plant as a rich source of phytochemical compounds.

CONCLUSION

The present study reveals that the stem, leaves and fruits of *Solanum nigrum* have potential sources of useful drugs due to the presence of pytochemical constituents and provides the information respect of their identification, strandadization of herbal drugs of folk medicinal practice of present era.

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REFERENCES

1. Farnsworth NR, Aereele O, Bingel AS. Medicinal plants in therapy. Bulletin of the world Health Organisation., 1985; 63: 965–981.
2. Karthikeyan A, Shanthi V, Nagasathaya A. A Preliminary Phytochemical and Antibacterial Screening of crude extract of the leaf of *Adhatoda vasica* L. Int. J. Green Pharm, 2009; 3: 78–80.

3. Okwu DE. Phytochemical and Vitamins content of indigeneous spices of South Eastern Nigeria. *J. Sustain, Agric, Environ.*, 2004; 6: 3–34.
4. Aktar MS, Munir. Evaluation of the Gastric and Anti – Ulcerogenic effects of *Solanum nigrum*, *Brassica oleraceae* and *Ocimum basilicum* in rats. *Journal of Ethanopharmacology.*, 2008; 27(1): 163-176.
5. Gogoi P, Islam M. Ethano medicinal Study of *Solanum nigrum* L. and *S.Myriancanthus* Dunal used by tribals and Non tribals from the districts of Upper Assam, India. *Asian. J. Exp, Bio, Sci.*, 2012; 3(1): 73–81.
6. Fluck. Medicinal plants and their uses. Newyork; W. Feulshom. com. Ltd., 1973; 7-15.
7. Sodipo OA, Akinjii JA, Ogunbamose JU. Studies on certain characteristic of extracts of bark of *Paninystalia macruras* (k.Schemp) pierre Exbeile. *Global. J. Pune. Appl. Sci.*, 2000; 83 -87.
8. Herman Janice R. “Protein and the body “Oklahoma cooperative Extension Service and Natural Resouces. Oklahoma state University., T – 3136 -1- T- 3163 -4.
9. Arya V, Gupta R, *Journal of Chemical and Pharmaceutical Research.*, 2011; 3(3): 447–456.
10. Ludwig DS, Pereira MA, Kroenke CH, Hilner JE, Van Horn L, Slattery ML, Jacobs DR. Dietary fiber, Weight gain, and Cardiovascular risk factors in young adults *J Am. Med. Assoc.*, 1999; 282: 1539-1546.
11. Pratt RJ, Chase CR. Fluorescence of powdered vegetable drug with particular reference to development of a system of identification. *J Am Pharm Assoc.*, 38: 324-33.
12. Harborne JB. Pytochemical methods (London: Champan and Hall Ltd., 1989; 19–21.
13. Trease GE, Evans WC. A Textbook of pharmacy 13 London : Bacillere Tinall Ltd, 1989; 19-21.
14. Oguyemi AO. In Sofowara A.ed Proceedings of a conference on African Medicinal Plants, Ife-Ife Univ Ife., 1979; 20-22.
15. Jackson BP, Snowdown DW. Powdered vegetable drugs, cheer Chil Ltd: London., 1968; 25.
16. Kumar D, Kumar K, Kumar S, Kumar T, Kumar A, Prakash O. *Asian Pacific jounal of Tropic Biomedicine.*, 2012; 169–175.
17. Arya V, Gupta R *Journal of chemical and Pharmaceutical Research.*, 2011; 3(3): 447-456.
18. Atanu FO, Ebiloma UG 1and Ajay E.I. Department of Biochemistry, Orogbonigeria. *Biotechnology and Molecular biology review.*, 2011; 6(1): 001-007.

19. Parvathi A, Gnana sundari S, Rekha S. Phytochemical evaluation of three species of *Solanum* L. Herbal study centre, Department of Botany, Holy cross college, Trichy, Tamil Nadu, India., 2013.
20. Kumawat NS, Chandhari SP, Wani NS, Deshmukh TA, Patil VR. Antidiabetic activity of ethanol extract of *Colocasia esculenta* leaves in alloxan induced diabetic rats. *Inter. J. Pharmatech Res.*, 2010; 2(2): 1246-1249.
21. Catopano AL. Antioxidant effects of flavonoids. *Angiol.*, 1997; 48: 39-46.
22. Kameshwararao B, Giri R, Kesavulu MM, Apparao C. Herbal medicine, In the treatment of diabetes mellitus, *Manphar Vaidya Patrika*, 1997; 1: 33-35.
23. Akthar MS, Munir M .Evaluation of Anti-ulcerogenic effect of *Solanum nigrum*, *Brassica oleraceae* and *Ocimum basilicum* in rats. *J. Ethanopharmacol.*, 1989; 27: 163-172.
24. Kritikar KR, Basu BD. *Indian medicinal palnts* 2nd Edn. Lalit Mohan Babu, Allahabad. 1935.
25. Cham BE, Wilson L. HPLC Sodomaceum of glycoalkaloid from *Solanum* plant med., 1987; 53: 59-62.
26. Cooper M R, Johnson AW. *Poisonous plants in Britain and their effects on animals and man.* London: H.M.S.O, 1984; 219-220.
27. Ansari SH, *Essentials of Pharmacognosy* 1st Edn. Birla Publication Pvt. Ltd. New Delhi. 2006.