

Phytochemical and Nutritional Evaluation of *Solanum diphylum* L.: A Potential Medicinal Plant

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Abstract

Solanum diphylum L., Solanaceae, is a medicinal shrub used extensively in traditional Indian medicine for the dealing of intestinal parasites, respiratory conditions, and skin infections. Phytochemical investigations reveal that *Solanum diphylum* L. contains diverse secondary metabolites, including steroidal alkaloids (glycoalkaloids), flavonoids, terpenoids, tannins, saponins, and other phenolic compounds. This chemical diversity underpins a broad spectrum of pharmacological activities. Extracts of *Solanum diphylum* L. have demonstrated anti-inflammatory, antioxidant, anticancer, anti-ulcer, antimicrobial, and anti-infective effects, in addition to enzyme-inhibitory (anti- α -amylase and anti- α -glucosidase) and immuno-modulatory properties. Alkaloids and phenols compounds were quantified because to their high concentration and significant pharmacological relevance. Their analysis underscores the therapeutic potential of *Solanum diphylum* L. in phytomedicine. Quantification study of total phenols and alkaloids reveals marked variations among plant parts. The leaf extract exhibits a total phenolic content of 73.88 mg of GAE per g of sample, compared to 11.82 mg of GAE per g of sample in stem and 8.76 mg of GAE per g of sample in root. Likewise, the total alkaloid content is highest in the leaf (4.34 ± 0.22 g %), followed by stem (3.01 ± 0.17 g %) and root (0.92 ± 0.037 g %). Proximate composition was also determined, including moisture, ash and fiber content. This data reflecting its water, fiber and inorganic content (mineral); contributing to the understanding of its overall health benefits.

Keywords: *Solanum diphylum* L., phytochemical analysis, phenols, alkaloids, proximate composition

Introduction

The Solanaceae family, commonly referred to as the nightshade family. The

globally distributed *Solanum* genus, documented as one of the largest and most diverse within the Solanaceae family, encompasses 1500A1800 species, securing its position among the top ten genera with the highest species richness in flowering plants (Frodin, 2004; S Basu et al.,2014). Members of Solanaceae play crucial roles in human diet, medicinal applications, and cultural traditions

Solanum diphyllum L. is a species of the Solanaceae family. This plant is known by various common and local names, including twinleaf nightshade, two-leaf nightshade, and tomatillo in Mexico (S Knapp,2002). *Solanum diphyllum* L. is indigenous to Mexico and Central America, also found in various tropical and subtropical countries. It is valued for its foliage and is a perennial shrub typically reaching heights of 1 to 2 meters. The plant is recognized by its smooth, lance-shaped leaves and bears spherical berries as fruits (FA Hamada et al.,2010). The plant's ability to do well in different environments makes it an interesting topic for ecological studies. The World Health Organization (WHO) states that an important portion of the global population relies on traditional medicines as their main form of healthcare. Medicinal plants are considered crucial for the expansion of life-saving medications. Secondary metabolites found in plants show various biological properties, comprising antioxidant, anti-apoptotic, anti-aging, anti-carcinogenic, anti-inflammatory, anti-atherosclerotic, cardiovascular protective, angiogenesis inhibition, and cell proliferation activity (DS Fabricant & NR Farnsworth, 2001). *Solanum diphyllum* L. shows anti-inflammatory, anticancer, antiulcer, antioxidant, antimicrobial, antiinfection, anti-alpha amylase, anti-alpha glucosidase, and immuno-modulatory activities and also exhibited encouraging cytotoxic effects on colon and breast cancer cell lines, which demonstrates its potential as an important reservoir of anticancer compounds (MA El-Sayed et al.,2009).

Objectives

- To conduct a comprehensive phytochemical screening of *Solanum diphyllum* L. using solvent extracts derived from methanol, n-hexane, ethyl acetate, and water.
- To quantitatively determine the concentrations of key secondary metabolites, specifically total phenols and total alkaloids
- To evaluate the fundamental composition of *Solanum diphyllum* L. by assessing percentage of moisture, overall ash composition, fiber content, acid-insoluble ash, and water-soluble ash, thereby elucidating its nutritional and physicochemical properties.

Data and Methodology

Collection plant material

Plant material of *Solanum diphyllum* L. was obtained from Hanging Garden located in Simla Nagar, Malabar Hill, Mumbai, Maharashtra (N 18° 57' 25.596", E 72° 48' 17.2872"). The plant material taxonomically Classified and confirmed in the Blatter herbarium at the Department of Botany by Dr. Pravin V. Kale, St. Xavier's college, Mumbai. The entire plant sample was dehydrated in exposure to air in the shade at ambient temperature and subsequently subjected to an oven at 40°C to eliminate moisture. The dried plant was then finely grounded, sieved and stored in airtight containers.

Phytochemical profiling

Phytochemical profiling of *Solanum diphyllum* L. involved using established methods to detect various bioactive compounds. Different tests were conducted to examine the phytochemical content of extracts obtained from methanol, n-hexane, ethyl acetate, and water. Solvents were chosen according to their ability to extract efficiently and their polarity to maximize yield. The tests aimed to identify Sugars, peptides, lipids, and amino acids, Steroids, Glycosides, Terpenoids, Saponin, Flavonoids, Alkaloids, Phenols, and Tannins within the plant material (RNS Yadav et.al.,2011; Ayoola et al.,2008; Harborne,1998).

Evaluation of carbohydrates

Benedict's examination

A reddish-brown precipitate formed when the extract was heated with two milliliters of Benedict's reagent, indicating the presence of carbohydrates.

Test of Fehling solutions

One milliliter of Fehling's solutions A and B were added to two milliliters of the test solution. After that, the mixture was boiled for five to ten minutes. Carbohydrates were present because a brick-red precipitate formed.

Evaluation of amino acids

Test for ninhydrin

Two milliliters of a 0.2% ninhydrin solution were used to boil the extract. The presence of amino acids was indicated by the appearance of a violet color.

Evaluation of lipids

Sudan 4 test

A few drops of Sudan IV were added to the test solution, shaken thoroughly, and then water was added. Lipids were present because a reddish-stained oil layer

formed.

Evaluation of protein

Xanthoproteic examination

A few drops of concentrated nitric acid (HNO_3) were added to the extract. Proteins were present when a yellow tint appeared.

Evaluation of tannins

Braymer's examination

A few drops of a 1% ferric chloride (FeCl_3) solution were added to the extract. Tannins were present when a blue or greenish tint appeared.

Evaluation of alkaloids

Wagner's examination

A few drops of Wagner's reagent (iodine solution in potassium iodide) were added to the extract. Alkaloids were present because a reddish-brown precipitate formed.

The Dragendroff test

A few drops of Dragendroff's reagent were added to the extract. Alkaloids were present because a reddish-brown precipitate formed.

Evaluation of glycosides

The Liebermann test

After combining the extract with acetic acid and chloroform, concentrated H_2SO_4 was added along the side of the test tube. Glycosides were indicated by a blue-green ring at the junction.

The Keller-Kiliani test

The extract was subjected to concentrated H_2SO_4 after being treated with glacial acetic acid and ferric chloride in the Keller-Kiliani test. The presence of glycosides was indicated by a reddish-brown ring at the interface.

Evaluation of saponin

Test for foam

Water was used to vigorously shake the extract. Saponins were present because of the persistent foam formation.

Evaluation of flavonoids

Test with an alkaline reagent

Indicating flavonoids, the extract turned yellow when NaOH was added and turned colorless when diluted acid was added.

Test for lead acetate

Lead acetate solution was used to treat the extract. Flavonoids were present because a yellow precipitate formed.

Evaluation of terpenoids

Salkowski's examination

Salkowski's test involved combining the extract with concentrated H_2SO_4 and chloroform. Steroids were visible at the interface as a reddish-brown tint.

Evaluation of phenols

Test for ferric chloride

A few drops of a 10% FeCl_3 solution were added to the extract. Phenolic compounds were present when a blue, green, or purple hue appeared

Evaluation of steroids

The Burchard test by Liebermann

Acetic anhydride and concentrated H_2SO_4 were used to treat the extract. Steroids or sterols were present when a blue-green tint developed.

Estimation of Phenols

The Folin-Ciocalteu reagent, as outlined by Slinkard and Singleton (1977), was used to measure the concentration of total phenolics in water extracts. Gallic acid was used as the external calibration standard. The solution was made by vigorously mixing 0.1 ml of the extract with 0.8 ml of DW and 0.1 ml of Folin-Ciocalteu reagent. Three milliliters of 2% Na_2CO_3 were added after three minutes of shaking. After that, the mixture was left to stand for two hours at room temperature. A UV-VIS spectrophotometer was used to measure the absorbance at 760 nm. Using a calibration curve derived from Gallic acid, the total phenolic content was expressed as Gallic acid equivalents.

Estimation of Alkaloids

The gravimetric method was used to identify the alkaloids present in the leaf, stem, and root. One liter of 10% acetic acid in ethanol was added to a 500-milliliter conical flask containing a 200-gram sample of plant material. Before being filtered, the mixture was covered and allowed to stand for four hours. To precipitate the alkaloids, concentrated aqueous ammonium hydroxide was added drop wise to the filtrate, which had been concentrated to a quarter of its initial volume on a water bath set at 60°C . After centrifuging the mixture, the precipitate was gathered and cleaned with 15% ammonium hydroxide. After 30

minutes of oven drying at 60°C, the alkaloid precipitate was weighed. According to Nimenibo-Uadia et al. (2017), the alkaloid content was computed as a percentage of the initial sample weight.

Proximate analysis

Ayurvedic pharmacopeia methods were used to determine the proximate composition in order to determine moisture, crude fiber, total ash, acid insoluble ash, water soluble ash, and total solid content.

Results and Discussion

One essential technique for locating bioactive substances is phytochemical analysis. It is a quick, affordable, and simple method for identifying different classes of phytochemicals found in plants. Phytochemicals indicate that a plant may hold promise as a potential reservoir of precursor compounds for drug development (GA Ayoola et al., 2008). Plant synthesizes a large range of metabolites, both primary and secondary with different functional groups. Phytochemicals suggests both physiological and medicinal activities. The qualitative phytochemical profiling of *Solanum diphyllum* L. was studied as mentioned in Table.1. (Here, PP = moderately to high, P = Low, A = absent or negligible, L=Leaf, S= Stem, R=Root)

Sr.No.	Primary & Secondary metabolites	Phytochemical tests	Water			Methanol			n Hexane			Ethyl acetate		
			L	S	R	L	S	R	L	S	R	L	S	R
1	Carbohydrates	Fehling's sol.test	P	P	P	P	P	A	P	A	A	A	A	A
		Benedict's test	P	A	P	P	A	A	A	A	A	A	A	A
2	Amino acids	Ninhydrin test	P	P	P	P	A	A	A	A	A	A	A	A
3	Lipids	Sudan 4 test	A	A	A	A	A	A	A	A	A	A	A	A
4	Proteins	Xenthoproteic test	A	A	A	A	A	A	A	A	A	A	A	A
5	Tannins	Braymer's test	P	A	A	P	A	A	A	A	A	A	A	A
6	Alkaloids	Dragendroff's test	P P	P	P	P P	P	A	P	A	A	P	A	A
		Wagner's reagent	P P	A	P	P	P	A	P	A	P	A	A	P

7	Glycosides	Liebemann's test	A	A	A	P	P	A	A	A	A	A	A	A
		KellerKiliani test	P	A	A	A	A	A	P	A	A	A	A	A
8	Saponin	Foam test	P	P	A	A	A	A	A	A	A	A	A	A
9	Flavonoids	Lead acetate test	P	P	A	A	P	P	A	A	A	A	A	A
		Alkline reagent test	P	P	P	A	P	P	A	A	A	A	A	A
10	Terpenoids	Salkowski's test	P	A	A	A	A	A	A	A	A	A	A	A
11	Steroids	Liebermann's Burchard	A	A	A	A	A	A	A	A	A	A	A	A
12	Phenols	Ferric chloride test	A	A	A	P P	P	A	A	A	A	P	A	A

Table.1: Phytochemical profiling of *Solanum diphyllum* L.

Phytochemical profiling of the plant samples showed that water and methanol extracts had more primary and secondary metabolites than hexane and ethyl acetate.

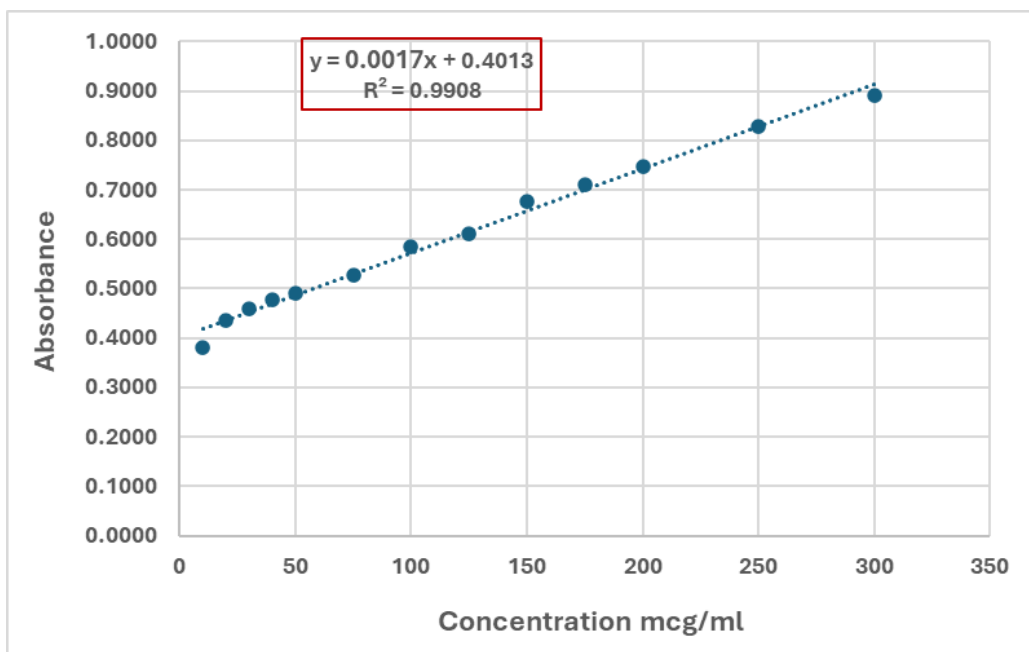
Sample	y value	Regression formula	Conc. in 50ml/gm extract (mg)
Leaf	0.6525	$y = 0.0017x \text{ P } 0.4013$	73.88
Stem	0.4415		11.82
Root	0.3715		8.764

Table.2: Estimation of phenols of *Solanum diphyllum* L.

Phenolic compounds are critical secondary metabolites in plants, known for their anti-hyperglycemic, anti-inflammatory properties and inhibit the development of diabetes. Since phenols have been shown to donate hydrogen atoms or electrons, neutralize free radicals, and prevent oxidative stress, the higher phenolic content is consistent with improved antioxidant activity (MA El-Sayed et al.,2009). A significant range in phenolic concentrations was found in the most recent quantitative assessment of *Solanum diphyllum* L.'s total phenol content using Folin-Ciocalteu reagent assays. The leaves had the highest phenolic content of

any part examined, followed by the stem and the root.

Figure 1: Calibration curve of Gallic acid



The standard curve for measurement, shown in Figure 1, follows the regression equation $y = 0.0017x + 0.4013$ with an R^2 value of 0.9908, indicating a strong linear relationship. This standard curve equation is represented as $y = mx + c$, where $m = 0.0017$ and $c = 0.4013$.

Sample	Calculation formula	Alkaloids (g %)
Leaf	$(\text{Weight of Alkaloid} / \text{Weight of Sample}) \times 100 \%$	4.34%
Stem		3.10%
Root		0.92%

Table.3: Estimation of alkaloids of *Solanum diphyllum* L.

Solanum diphyllum L. contains alkaloids with a variety of pharmacological properties, such as analgesic, antibacterial, and anticancer effects. These bioactive substances help to reduce oxidative stress, induce cytotoxic reactions in cancerous cells, inhibit the growth of harmful microorganisms, and lessen inflammatory and nociceptive disorders (MA El-Sayed et al.,2009). Results obtained from the quantitative evaluation of alkaloids (Table 3) showed the leaf has the highest percentage of alkaloids, the stem and the root having lower levels.

Sr. no	Experimental Studies	Observations for powdered of <i>Solanum diphyllum</i> L.		
		Leaf	Stem	Root
1.	Total ash value	14.05±0.18	8.66±0.23	7.41±0.23
2.	Acid insoluble ash value	0.32±0.06	0.40±0.02	0.88±0.29
3.	Water soluble ash value	3.97±0.29	3.28±0.2	1.24±0.16
4.	Moisture content	78.63±0.94	63.32±2.24	56.82±0.35
5.	Total solid content	21.37±0.06	36.68±1.23	43.18±0.65
6.	Crude fiber content	10.67±0.09	19.89±0.34	31.2±0.25

Table.3: proximate composition of *Solanum diphyllum* L.

The proximate composition of *Solanum diphyllum* L. (leaf, stem, and root) reveals significant variations in moisture, ash, and solid content. The leaf has the highest moisture content, followed by the stem, with the root showing the lowest moisture. This suggests that the root is less prone to microbial spoilage if properly dried. The leaf has the highest total ash content, indicating a higher concentration of minerals, while the root contains a moderate level. The acid-insoluble ash is highest in the root, suggesting stronger mineral resistance to digestion. Water-soluble ash is highest in the leaf, which may enhance nutrient bioavailability. The root has the highest total solid content, indicating greater solid material for therapeutic use. The root's high fiber is likely beneficial for digestive health, weight management, and prevention of diseases such as cancer, diabetes, and cardiovascular disorders. The composition of *Solanum diphyllum* L. highlights its potential nutritional and therapeutic value.

Conclusion

The phytochemical investigation revealed the presence of carbohydrates, alkaloids, tannins, phenols, terpenoids, saponin and flavonoids bioactive compounds recognized for their diverse physiochemical and pharmacological effects. Their presence supports the traditional medicinal use of *Solanum diphyllum* L. in managing various health conditions. In addition to confirming conventional applications, this thorough biochemical profiling offers a strong basis for upcoming studies that seek to identify active ingredients and comprehend their modes of action in order to possibly develop pharmaceuticals. Additionally, proximate analysis highlighted significant nutritional properties, including high moisture, ash, and fiber content, which may enhance the plant's

therapeutic efficacy.

References

1. Ayoola, G. A., Coker, H. A., Adesegun, S. A., Adepoju-Bello, A. A., Obaweya, K., Ezennia, E. C., & Atangbayila, T. O. (2008). Phytochemical screening and antioxidant activities of some selected medicinal plants used for malaria therapy in Southwestern Nigeria. *Tropical journal of pharmaceutical research*, 7(3), 1019-1024.
2. Basu, S., Zandi, P., Cetzal-Ix, W., & Sengupta, R. (2014). Solanaceae: The nightshade family. *The Encyclopedia of Earth*, 1-20.
3. El-Sayed, M. A., Mohamed, A. E. H. H., Hassan, M. K., Hegazy, M. E. F., Hossain, S. J., Sheded, M. G., & Oht, S. (2009). Cytotoxicity of 3-O-(β -D-glucopyranosyl) etioline, a steroidal alkaloid from *Solanum diphyllum* L. *Zeitschrift für Naturforschung C*, 64(9-10), 644-649.
4. Fabricant, D. S., & Farnsworth, N. R. (2001). The value of plants used in traditional medicine for drug discovery. *Environmental health perspectives*, 109(suppl 1), 69-75.
5. Frodin, D. G. (2004). History and concepts of big plant genera. *Taxon*, 53(3), 753-776.
6. Hamada, F. A., Hamed, A. I., Sheded, M. G., & Shaheen, A. M. (2010). Macro, micro-morphological and bioactivity aspects of naturalized exotic *Solanum diphyllum* L. *Al-Azhar Bulltein of Science. (ISCAZ 2010)*, 175-206.
7. Harborne, A. J. (1998). *Phytochemical methods a guide to modern techniques of plant analysis*. springer science & business media
8. Knapp, S. 2002. *Solanum* section *Geminata* (Solanaceae). *Fl. Neotrop. Monogr.* 84: 1–404.
9. Nimenibo-Uadia, R., Ugwu, I., Erameh, T., & Osunde, E. (2017). Estimation of tannins, alkaloids, saponins and proximate composition of *Vernonia amygdalina* (Del) root. *International Journal of Herbal Medicine*, 5(3), 88-92.
10. Payyappallimana, U. (2008). *Ayurvedic Pharmacopoeia. Modern and Global Ayurveda: Pluralism and Paradigms*, 139.
11. Slinkard, K., & Singleton, V. L. (1977). Total phenol analysis: automation and comparison with manual methods. *American journal of enology and viticulture*, 28(1), 49-55.
12. Yadav, R. N. S., & Agarwala, M. (2011). Phytochemical analysis of some medicinal plants. *Journal of phytology*, 3(12).