RESEARCH PAPER

Phytochemical Investigation and Anti Microbial Activity of Allamanda Cathartica

Praveen Kumar Dasari[1]*, Ch. Supriya[2], Ch. Nagabhushanam[2], Rajaboina Lakshmi[1], Krishna Durga Peddini[1], Nandhikola Harika[1], Bellamkonda Vishnu Vardhan[1] and Gundepuri Anil[1]

[1]Department of Pharmaceutical Biotechnology, Mother Teresa Pharmacy College Sathupally, Khammam, Telangana, India
[2]Department of Pharmacology, KVSR Siddhartha College of Pharmaceutical Sciences, NTR District, Andhra Pradesh, India
drdppharma@gmail.com

Received: 24th April, 2026; Revised: 20th May 2026; Accepted: 30th June, 2026; Available Online: 10th July, 2026

ABSTRACT

The exploration of medicinal plants as alternative sources of antimicrobial agents has gained significant attention due to the increasing emergence of drug-resistant microorganisms. *Allamanda cathartica*, a medicinal plant widely distributed in tropical regions, is known for its diverse therapeutic properties attributed to the presence of various bioactive phytochemicals. The present study aimed to investigate the phytochemical constituents and evaluate the antimicrobial potential of leaf extracts of *A. cathartica*. Fresh leaves of *A. cathartica* were collected, processed, and extracted using suitable solvents under laboratory conditions. Preliminary phytochemical screening was carried out using standard qualitative methods to identify the presence of major secondary metabolites. The antimicrobial activity of the extracts was assessed against selected microorganisms isolated from soil samples using agar well diffusion and minimum inhibitory concentration (MIC) techniques. Different concentrations of the plant extract were prepared to determine their effectiveness in inhibiting microbial growth. Phytochemical analysis revealed the presence of several bioactive compounds, including alkaloids, flavonoids, tannins, saponins, phenolic compounds, glycosides, and terpenoids. The antimicrobial evaluation demonstrated that the leaf extracts exhibited inhibitory activity against microorganisms. The degree of inhibition increased with increasing extract concentration, indicating a concentration-dependent antimicrobial effect. The results suggest that *A. cathartica* possesses significant antimicrobial potential and could serve as a valuable source of natural bioactive compounds for pharmaceutical applications.

Keywords: *Allamanda cathartica*, phytochemical investigation, antimicrobial activity, medicinal plant, leaf extract, bacterial isolates.

How to cite this article: Dasari PK, Supriya C, Nagabhushanam C, Lakshmi R, Peddini KD, Harika N, Vishnu Vardhan B, Anil G, Phytochemical Investigation and Anti Microbial Activity of Allamanda Cathartica. Int J Drug Deliv Technol. 2026;16(6): 636-641. DOI: 10.25258/ijddt.16.6.100
Source of support: Nil.
Conflict of interest: None

INTRODUCTION

Medicinal plants have been utilized for centuries as a primary source of therapeutic agents for the prevention and treatment of various diseases. The growing incidence of antimicrobial resistance and the reduced effectiveness of conventional antibiotics have encouraged researchers to explore natural products as alternative sources of antimicrobial compounds. Plant-derived bioactive substances are known to possess diverse pharmacological properties, including antimicrobial, antioxidant, anti-inflammatory, and wound-healing activities. Consequently, the investigation of medicinal plants has become an important area of pharmaceutical and microbiological research. *Allamanda cathartica*, commonly known as Golden Trumpet, belongs to the family Apocynaceae and is widely cultivated as an ornamental plant in tropical and subtropical regions. In traditional medicine, various parts of the plant have been used for the management of ailments such as skin infections, jaundice, malaria, inflammation, and digestive disorders. The medicinal properties of *A. cathartica* are attributed to the presence of several biologically active phytochemicals, including alkaloids, flavonoids, tannins, saponins, terpenoids, glycosides, and phenolic compounds.

These compounds have attracted considerable scientific interest due to their potential therapeutic applications. Phytochemical investigation serves as an essential step in identifying the chemical constituents responsible for the biological activities of medicinal plants. Understanding the phytochemical profile of a plant can provide valuable insights into its pharmacological potential and facilitate the discovery of novel therapeutic agents. The increasing prevalence of multidrug-resistant microbial strains has

**Author for Correspondence: drdppharma@gmail.com*

become a major global health concern. Therefore, there is an urgent need to identify new antimicrobial agents that are effective, affordable, and environmentally friendly. Plant extracts have emerged as promising alternatives because they contain multiple bioactive compounds capable of inhibiting microbial growth through various mechanisms. The present study was undertaken to investigate the phytochemical constituents and evaluate the antimicrobial activity of *Allamanda cathartica* leaf extracts against selected bacterial and fungal isolates. Preliminary phytochemical screening was performed to detect the presence of major secondary metabolites, while antimicrobial activity was assessed using standard microbiological techniques. The findings of this study may contribute to the development of plant-based antimicrobial agents and provide scientific evidence supporting the medicinal value of *A. cathartica*.

PLANT PROFILE

Common Names: Golden Trumpet**,**Yellow Bell**,** Golden Bell**,** Common Allamanda

Botanical Description: *Allamanda cathartica* is a perennial evergreen shrub or climbing vine belonging to the family Apocynaceae. It is native to South and Central America and is widely cultivated in tropical and subtropical regions as an ornamental plant due to its attractive bright yellow flowers. The plant grows rapidly and can attain a height of 2–6 meters under favorable conditions. The leaves are simple, glossy, dark green, and arranged in whorls of three or four. They are lanceolate to elliptic in shape with smooth margins and pointed tips. The flowers are large, trumpet-shaped, bright yellow in color, and bloom throughout most of the year. The fruit is a spiny capsule containing numerous seeds.

Significance of the Plant

The rich phytochemical composition of *Allamanda cathartica* makes it a valuable medicinal plant with considerable pharmaceutical potential. Its antimicrobial properties suggest that it may serve as a natural source of bioactive compounds for the development of alternative therapeutic agents against microbial infections. Consequently, phytochemical investigation and antimicrobial evaluation of this plant are important for validating its traditional uses and exploring its future applications in drug discovery.

Fig 01: *Allamanda cathartica* plant

Fig 02: *Allamanda cathartica* flower

MATERIALS AND METHODS

Collection of Plant Material

Fresh and healthy leaves of *Allamanda cathartica* were collected from the local area of Sathupally. The collected plant material was free from disease and physical damage. The samples were placed in clean polyethylene bags and transported to the laboratory for further processing.

Identification and Authentication of Plant Material

The collected plant specimen was identified and authenticated based on its morphological characteristics with the help of a qualified botanist from the Department of Botany. The authenticated plant material was used for phytochemical and antimicrobial studies.

Preparation of Plant Material

The collected leaves were washed thoroughly with tap water followed by distilled water to remove dust and other contaminants. The leaves were shade-dried at room temperature for 10–15 days until a constant weight was obtained. The powdered material was stored in airtight containers for further extraction.

Extraction of Plant Material by Soxhlet Extraction

The dried leaf powder was subjected to Soxhlet extraction using different solvents of varying polarity, namely ethanol, methanol, petroleum ether, and chloroform. Each solvent was used separately, and the extraction was continued for 6–8 hours until exhaustive extraction was achieved.. The obtained extracts were concentrated using a rotary evaporator and further dried at room temperature to remove residual solvent. The dried extracts were weighed, transferred into sterile containers, and stored at 4°C until further analysis.

Preparation of Extract Solutions

Stock solutions of the crude extracts were prepared by dissolving a known quantity of each extract in an appropriate solvent. Different concentrations of the extracts were prepared for phytochemical screening and antimicrobial studies.

Preliminary Phytochemical Screening

The ethanol, methanol, petroleum ether, and chloroform extracts were subjected to preliminary phytochemical screening using standard qualitative methods. The extracts were tested for the presence of various secondary metabolites such as alkaloids, flavonoids, tannins, saponins, phenolic compounds, glycosides, terpenoids, steroids, carbohydrates, and proteins.

Preliminary Phytochemical Screening of *Allamanda cathartica* Leaf Extracts

Screening for carbohydrates

Molisch test: To 2 ml of extract add 3 drops of α-naphthol (20% in ethanol), then 1ml of concentrated sulphuric acid was added by side of the test tube. Reddish-violet ring was noticed at the junction of the two layers indicated the presence of carbohydrates.

Benedict's test: To 2 ml of extract add few drops of Benedict's reagent, heat it for five minutes dark red precipitate is observe indicates the presence of carbohydrates..

Screening for glycosides

Keller killiani test: To 2 ml of extract add 0.5 mL of glacial acetic acid and 2-3 drops of ferric chloride then add 1 mL of concentrated H_2SO_4 on the walls of the test tube. Deep blue colour was noticed at the junction of two solutions indicates the presence of cardiac glycosides.

Screening for alkaloids

(a) Dragendorff's test: To 2 mL of extract add 1 mL of Dragendorff"s reagent, orange red precipitate was noticed that indicates the presence of alkaloids.

(b) Hager's test: To 2mL of extract add few drops of Hager's reagent, yellow precipitate was noticed that indicates the presence of alkaloids.

Screening for terpenoids

Salkowski test: To 2 ml of extract add 2 ml of chloroform, then add of 3 ml concentrated sulfuric acid (H_2SO_4). Reddish brown color was noticed that indicates the presence ofterpenoids .

Screening for steroids

Liebermann burchard test: To sample extract add chloroform and filter, to the filtrate add few drops of acetic anhydride, heat and cool it. Add H_2SO_4, brown ring was obtained .

Screening for tannins

Ferric chloride test: To 2 ml of extract add a few drops of 10% Ferric chloride solution, blackish blue colour indicates the presence of gallic tannins and a green-blackish colour indicates the presence of catechol tannins.

Screening for flavonoids

Alkaline reagent test: To 2 ml of extract add three drops of sodium hydroxide. Initially, it shows a deep yellow colour and later it becomes colorless by the adding few drops of dilute hydrochloric acid, indicates the presence of flavonoids.

Screening for phenols

2 ml of extract add 5ml of distilled water and few drops of 10% ferric chloride solution, a blue color indicates the presence of phenol compounds.

Tests for proteins

Biuret test: To 2 ml of extract add 2 drops of 3% copper sulphate and few drops of 10% sodium hydroxide, a violet or red colour was observed indicates the presence of proteins.

Ninhydrin test: To 2 ml of extract add 2 drops of 0.2% freshly prepared ninhydrin solution, purple colour indicates the presence of proteins.

Test for triterpenoids

Horizon test: To 2 ml of extract add 2 ml of trichloroacetic acid, red precipitate was noticed indicates the presence of terpenoids.

Determination of antimicrobial activity

The antimicrobial activity of *Allamanda cathartica* leaf extracts was evaluated against selected bacterial and fungal isolates using the agar well diffusion method. Nutrient Agar (NA) medium was used for bacterial cultures, while Potato Dextrose Agar (PDA) medium was used for fungal cultures. The media were prepared according to standard laboratory procedures and sterilized by autoclaving at 121°C for 15 minutes. The bacterial and fungal inocula were evenly spread over the surface of the respective sterile agar plates using sterile cotton swabs under aseptic conditions Different concentrations of ethanol, methanol, chloroform, and petroleum ether extracts of *Allamanda cathartica* leaves were prepared and introduced into the wells. The inoculated plates were incubated at 37°C for 24 hours for bacterial cultures and at 28°C for 48–72 hours for fungal cultures. Following incubation, the antimicrobial activity was assessed by measuring the diameter of the clear zones of inhibition surrounding the wells.The mean values of the inhibition zones were calculated and used for comparative evaluation of the antimicrobial effectiveness of the different solvent extracts.

Determination of minimum inhibitory concentration

The Minimum Inhibitory Concentration (MIC) of the leaf extracts was determined using the broth dilution method. Serial dilutions of each extract were prepared in sterile nutrient broth for bacterial isolates and potato dextrose broth for fungal isolates. A standardized microbial suspension was inoculated into each dilution and incubated under appropriate conditions. After incubation, the tubes were examined for visible microbial growth. The lowest concentration of the extract that completely inhibited visible growth of the test microorganism was recorded as the Minimum Inhibitory Concentration (MIC). The MIC values were used to assess the potency of the extracts and compare their antimicrobial efficacy against the selected microorganisms.

RESULTS AND DISCUSSION

Phytochemical Screening

Preliminary phytochemical screening of the leaf extracts of *Allamanda cathartica* revealed the presence of several biologically active secondary metabolites. The ethanol and methanol extracts showed the highest number of phytochemical constituents, including alkaloids, flavonoids, tannins, saponins, phenolic compounds, glycosides, terpenoids, steroids, carbohydrates, and proteins. Chloroform extract exhibited moderate phytochemical content, whereas petroleum ether extract contained mainly non-polar compounds such as terpenoids and steroids. The presence of these phytochemicals indicates that *A. cathartica* possesses significant medicinal potential. The variation in phytochemical composition among different solvent extracts may be attributed to differences in solvent polarity and extraction efficiency.

Table 01: Preliminary Phytochemical Screening of *Allamanda cathartica* Leaf Extracts

Phytochemical Constituents	Ethanol Extract	Methanol Extract	Chloroform Extract	Petroleum Ether Extract
Alkaloids	+	+	+	-
Flavonoids	+	+	-	-
Tannins	+	+	-	-
Saponins	+	+	-	-
Phenolic Compounds	+	+	+	-
Glycosides	+	+	+	-
Terpenoids	+	+	+	+
Steroids	+	+	+	+
Carbohydrates	+	+	-	-
Proteins	+	+	-	-

Antimicrobial Activity

The antimicrobial activity of the ethanol, methanol, chloroform, and petroleum ether extracts of *Allamanda cathartica* leaves was evaluated using the agar well diffusion method. The extracts exhibited varying degrees of inhibitory activity against the tested bacterial and fungal isolates. Among the extracts, ethanol and methanol demonstrated comparatively higher antimicrobial activity, as evidenced by larger zones of inhibition. Chloroform extract showed moderate activity, while petroleum ether extract displayed relatively lower antimicrobial effects. The enhanced antimicrobial activity observed in the ethanol and methanol extracts may be attributed to their ability to extract a wider range of bioactive compounds, particularly phenolics, flavonoids, tannins, and alkaloids. These compounds are known to inhibit microbial growth by disrupting cell membranes, interfering with enzyme activity, and affecting nucleic acid synthesis.

Table 02: Antimicrobial activity of *Allamanda cathartica* Leaf Extracts

Extract	Zone of Inhibition (mm)				
	25 µg/mL	**50 µg/mL**	**100 µg/mL**	**150 µg/mL**	**200 µg/mL**
Ethanol	4.2 ± 0.2	6.1 ± 0.3	8.4 ± 0.2	10.6 ± 0.3	10.9 ± 0.2
Methanol	3.9 ± 0.2	5.8 ± 0.2	7.9 ± 0.3	10.1 ± 0.2	10.6 ± 0.3
Chloroform	2.8 ± 0.1	4.5 ± 0.2	6.7 ± 0.2	8.9 ± 0.3	10.0 ± 0.2
Petroleum Ether	2.1 ± 0.1	3.8 ± 0.2	5.4 ± 0.2	7.2 ± 0.2	8.5 ± 0.3

CONCLUSION

The present study successfully investigated the phytochemical constituents and antimicrobial activity of *Allamanda cathartica* leaf extracts prepared using ethanol, methanol, chloroform, and petroleum ether as extraction solvents. Preliminary phytochemical screening revealed the presence of various bioactive compounds, including alkaloids, flavonoids, tannins, saponins, phenolic compounds, glycosides, terpenoids, and steroids, indicating the rich phytochemical composition of the plant. The antimicrobial evaluation demonstrated that the leaf extracts exhibited inhibitory activity against the tested bacterial and fungal isolates. The antimicrobial effect increased with increasing extract concentration, showing a concentration-dependent response. Among the four solvent extracts, ethanol and methanol extracts showed comparatively higher antimicrobial activity, which may be attributed to their ability to extract a greater number of biologically active phytochemicals. The findings of this study suggest that *Allamanda cathartica* possesses significant antimicrobial potential and can serve as a promising source of natural bioactive compounds for pharmaceutical and therapeutic applications. The results support the traditional medicinal use of the plant and highlight its potential role in the development of alternative antimicrobial agents to combat microbial infections and antimicrobial resistance.

ACKNOWLEDGEMENT

The authors express their sincere thanks to the management, Mother Teresa Pharmacy College, Sathupally, Telangana for providing the necessary facilities to carry out the research work.

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