

An Analysis of the Antimicrobial Activity of Methanol and Chloroform Extracts of *Cayratia trifolia*'s Leaf and Stem

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Abstract: Medicinal plants are widely used in Ayurveda for treating various ailments, and *Cayratia trifolia* is one such plant that exhibits broad medicinal properties. Known as Fox grape in English and Amlabel in Hindi, this plant belongs to the Vitaceae family. It contains various bioactive compounds that can be used directly or in extracted forms to manage different diseases. In this study, we investigated the antimicrobial potential of methanol and chloroform extracts from the stem and leaves of *Cayratia trifolia*. Our findings showed that methanol extracts from both the stem and leaves exhibited higher antibacterial and antifungal activity compared to their chloroform counterparts. *Cayratia trifolia* has been traditionally used for managing fever, rheumatic arthritis, and rheumatic fever, as well as for its anti-inflammatory and antimutagenic activity. Overall, these results suggest that *Cayratia trifolia* extracts could serve as natural sources of antimicrobial agents for treating bacterial and fungal infections.

Keywords: *Cayratia trifolia*; methanol; chloroform; bacteria; fungi.

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1. Introduction

Medicinal plants are very ancient and true natural medicines useful for treating different diseases. They can be used directly or in extracted forms for the management of various ailments due to the presence of various secondary metabolites [1]. The use of plants in the traditional remedy of many other cultures has been widely documented. These plant-based systems continue to play a significant role in health care, and it has been projected by the World Health Organization that around 80% of the 'world's inhabitants rely mainly on traditional medicines for their primary health care, although plant products also play a main role in the health care systems of the remaining 20% of the population mostly residing in developed countries [2,3]. Many plants contain a variety of phytopharmaceuticals, which have found very important applications in the fields of agriculture and human and veterinary medicine. Natural products play a dominant role in developing novel drugs, leading to the treatment and prevention of diseases [4,5]. The valuable medicinal properties of different plants are due to the presence of several antioxidants like saponins, tannins, alkaloids, alkenyl phenols, glycoalkaloids, flavonoids, sesquiterpenes lactones, and terpenoids [6]. Currently, synthetic antioxidants might be unsafe, and their toxicity has been criticized. It is generally assumed that frequent use of plant-derived phytochemicals may contribute to shifting the stability toward a sufficient antioxidant status. As a result, attention to natural antioxidants, particularly plant origin, has been deeply amplified in recent years [7].

In the present study, we evaluated the potential antimicrobial activity of *Cayratia trifolia* stem and leaf powder. Experimental plant *Cayratia trifolia* is considered a renowned folk medicine used against diabetes, arthritis, rheumatic pains, respiratory problems, wounds, urinary infections, dysentery, and also aphrodisiacs. Stem, leaves, and roots are reported to possess hydrocyanic acid and delphinidin. Several flavonoids, such as cyanidins, are reported in the leaves [8]. Infusion of seeds along with extract of tubers is traditionally given orally to diabetic patients to check blood sugar levels. The whole plant is used in diuretics, tumors, neuralgia, and splenopathy [9]. The paste of tubers is applied on the affected part to treat snake bites.

1.1. Taxonomical description.

Cayratia trifolia is a plant belonging to the family Vitaceae, which is part of the Plantae kingdom. The plant is categorized under the order Vitales and the genus Cayratia. The species name, trifolia, is derived from the Latin word meaning "three-leaved," which is a reference to the plant's characteristic leaves. The plant is commonly known as Fox grape in English and Amlabel in Hindi [10]. *Cayratia trifolia* is native to Asia and can be found in several countries, including India, Nepal, and Bhutan. The plant is a woody climber with tendrils that can grow up to 10 meters long. The plant leaves are trifoliate, meaning they are divided into three leaflets, and the flowers are small and greenish-white in color. *Cayratia trifolia* has been used in traditional medicine for treating a wide range of ailments, including fever, rheumatic arthritis, and rheumatic fever, and its anti-inflammatory and antimicrobial properties [11,12]. Understanding the taxonomy of plants like *Cayratia trifolia* is important for properly identifying, classifying, and studying their biological properties.

2. Materials and Methods

2.1. Sample collection.

The plant material was collected from the Ranthambore hills area (Sawai Madhopur), Rajasthan, and was authenticated in the Department of Botany, the University of Rajasthan. It was shaded dry and made powder form.

2.2. Sample preparation.

The crude extract was obtained by extraction. 20 gm of dried powder of the plant parts was macerated in 200 ml of respective organic chemical compounds (methanol and chloroform); it was kept in the rotatory soxhlet apparatus for 24 hours. In the next step, the extract was filtered and dried. It was stored in an airtight bottle at 40°C. The dried extract was further dissolved in the dimethyl sulphoxide (DMSO) solution for preparation of a final concentration 1 mg/ml and kept at 4°C until being used.

2.3. Isolates Inoculation

The agar well-diffusion method was followed to determine the antimicrobial activity. Nutrient Agar (NA) and Potato Dextrose Agar plates were swabbed (sterile cotton swabs) with 8-hour-old - broth cultures of respective bacterial and fungal isolates. Wells (10mm diameter and about 2 cm apart) were made in each of these plates using a sterile cork borer. A stock solution was prepared at a concentration of 1 mg/ml. Plant extract (methanol and chloroform)

sensitivity against different bacterial and fungal isolates in the volume of concentration loaded 40µl, 60µl, 80µl and 100µl into the wells and allowed to diffuse at room temperature for 2hrs. The plates were incubated at 37°C for 18-24 hours for bacterial pathogens and 28°C for 48 hours for fungal pathogens [13].

3. Results and Discussion

Antimicrobial activity is the ability to kill or inhibit disease-causing microorganisms, and it can be achieved using different types of antimicrobial agents. Such agents may exhibit either antibacterial or antifungal activity.

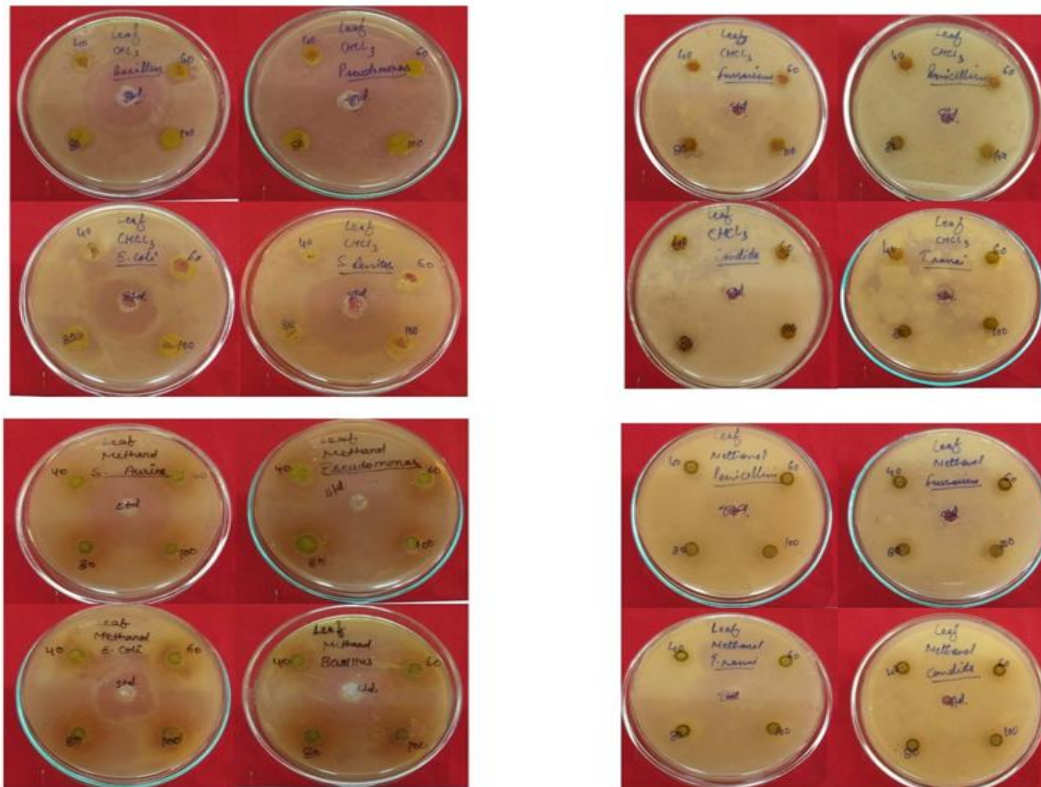


Figure 1. The antimicrobial activity of *Cayratia trifolia* leaf was investigated through chloroform and methanolic extracts. The chloroform extracts were labeled A and B, while the methanolic extracts were labeled C and D.

Figure 1 shows the antimicrobial activity of *Cayratia trifolia* leaf extracts on media plates. The antibacterial and antifungal activity of *Cayratia trifolia* leaf extracts (chloroform and methanol) is shown in Figure 2 and Figure 3. The bar graph in Figure 2 illustrates the antibacterial activity of methanol leaf extracts of *Cayratia trifolia* against four different bacterial strains. The results indicate that the methanol leaf extracts exhibit varying degrees of antibacterial activity against all tested bacterial strains, with the highest activity observed against *E. coli*. These findings suggest that *Cayratia trifolia*'s methanol leaf extracts possess potential antibacterial agents that could be useful in treating bacterial infections.

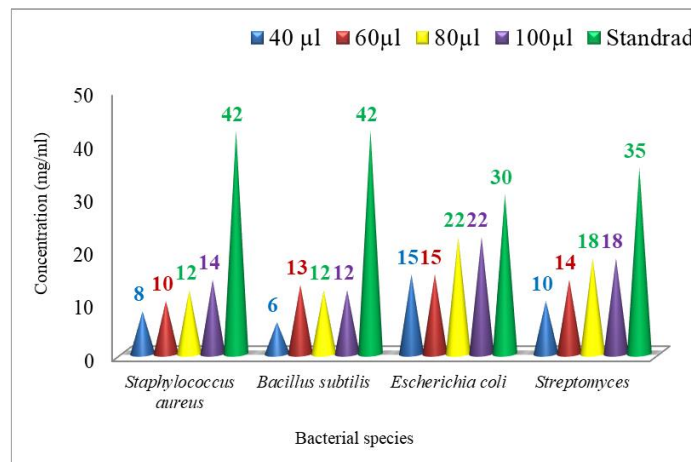


Figure 2. Antibacterial activity of methanol leaf extracts of *Cayratia trifolia* leaf. (standard: Ciprofloxacin 1mg/ml).

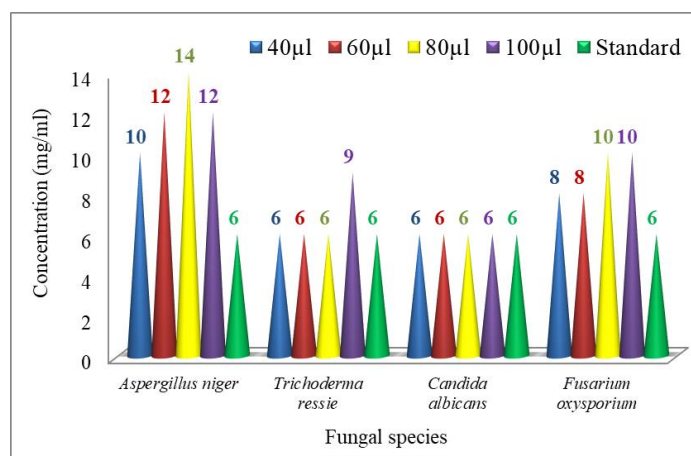


Figure 3. Antifungal activity of methanol leaf extracts of *Cayratia trifolia*. (Standard: Itraconazole 1mg/ml).

The bar graph in Figure 3 represents the antifungal activity of methanol leaf extracts of *Cayratia trifolia* against four different fungal strains, namely *Aspergillus niger*, *Trichoderma reessie*, *Candida albicans*, and *Fusarium oxysporium*. The results indicate that the methanol leaf extracts exhibit significant antifungal activity against all tested fungal strains, with the highest activity observed against *A. niger*. These findings suggest that *Cayratia trifolia*'s methanol leaf extracts possess potential antifungal agents that could be useful in treating fungal infections. The results also suggest that further investigations are warranted to isolate and identify the specific bioactive compounds responsible for the observed antifungal activity.

In this experiment, we investigated the antimicrobial activity of the leaf and stem extracts of *Cayratia trifolia*. The plant parts were shade-dried and extracted using methanol and chloroform. The reconstitution of plant compounds was done using 3 percent DMSO. Nutrient agar and Potato Dextrose Agar were used for bacterial and fungal activity, respectively.

The results showed that leaf and stem extracts exhibited significant antimicrobial activity against the tested microorganisms. The methanol extract of the leaf showed the highest antimicrobial activity against all tested microorganisms, followed by the chloroform extract of the leaf and the methanol extract of the stem. Other researchers have also obtained similar results [14,15]. However, the chloroform extract of the stem showed relatively low antimicrobial activity compared to other extracts. These findings are consistent with previous studies on *Cayratia trifolia*, which have reported the presence of various bioactive compounds,

such as flavonoids, phenols, alkaloids, and terpenoids, which have exhibited antimicrobial properties. For example, one study found that the flavonoids isolated from *Cayratia trifolia* exhibited significant antibacterial activity against *Staphylococcus aureus* and *Bacillus subtilis* [16-18]. The findings of this study are consistent with previous studies on *Cayratia trifolia*, which have reported the presence of various bioactive compounds, such as flavonoids, phenols, alkaloids, and terpenoids, which have exhibited antimicrobial properties [19,20]. Flavonoids, in particular, have been found to be the major bioactive compounds responsible for the observed antimicrobial activity of *Cayratia trifolia* [21]. Flavonoids are known to exhibit a wide range of pharmacological properties, including antimicrobial, antioxidant, anti-inflammatory, and anticancer activities [22-24]. The use of plant-based medicines has been a traditional practice for centuries and is still widely used in many parts of the world [25-27]. However, the scientific validation of the medicinal properties of plant-based remedies is essential for developing new drugs to treat various diseases [28]. The discovery of new antimicrobial compounds from medicinal plants could provide an alternative to conventional antibiotics, which are increasingly becoming ineffective due to the development of antibiotic-resistant bacteria [29,30]. The present study has important implications for identifying new sources of therapeutically and industrially valuable compounds from medicinal plants. The present study suggests that the leaf and stem extracts of *Cayratia trifolia* could be a potential source of new antimicrobial compounds that can be used to develop novel drugs for treating infectious diseases. However, further studies are needed to isolate and identify the active compounds responsible for the observed antimicrobial activity and investigate their action mechanism.

4. Conclusions

This study assessed the antimicrobial capabilities of methanol and chloroform extracts derived from *Cayratia trifolia*'s stems and leaves. The results reveal substantial antimicrobial activity in stem and leaf extracts, indicating their potential richness in bioactive compounds that could hold medicinal value. Our findings suggest that stem and leaf extracts contain bioactive compounds that could be valuable in medicine. Overall, the results support the potential use of *Cayratia trifolia* extracts as sources of natural antimicrobial agents that could be used for treating bacterial and fungal infections. Additionally, these extracts may have potential applications in pharmaceutical drug development for treating wounds and other related conditions.

In conclusion, this study provides evidence of the antimicrobial potential of *Cayratia trifolia* and suggests that the plant could be a potential source of new antimicrobial compounds. Further studies are needed to isolate and identify the bioactive compounds responsible for the observed antimicrobial activity and investigate their action mechanism. Subsequent investigations are warranted to meticulously isolate and characterize the precise bioactive constituents accountable for the observed antimicrobial efficacy. Furthermore, an in-depth exploration of these compounds is essential to unveil their potential therapeutic attributes, thereby expanding our understanding of their medicinal applicability.

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Conflicts of Interest

The authors declare no conflict of interest.

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