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# *In vitro* antimicrobial and Insecticidal activities of the methanolic extract of *Syzygium cumini*

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**Abstract**

The crude methanolic extract of *Syzygium cumini* was investigated for antibacterial and insecticidal biological activity. The test sample showed good antibacterial activity against Escherichia coli (20 mm), Staphylococcus aureus (19 mm), and Pseudomonas aeruginosa (18 mm), respectively. The Sample showed a significant zone of inhibition against Escherichia coli (25mm) and Salmonella typhi (22 mm), respectively. The test samples A, B, and C showed good insecticidal activity against *Tribolium castaneum* (58%), *Rhyzopertha Dominica* (55%), and *Callosbruchus analis* (60%), respectively.

**KEYWORDS.** *Syzygium cumini*, Antibacterial, Phytotoxic,Insecticidal Activity,

## 1.0 INTRODUCTION

There are an estimated 20,000 plants classified as medicinal that are used for therapy. Plants have become more interesting to modern medicine in recent years due to studies on therapeutic plants and the active ingredients obtained from them [1]. Many bioactive components found in herbal plants can be used to create medicines for a wide range of illnesses. The scientists estimated that there were about 391,000 different plant species, with flowering plants making up about 94% of the total. The Food and Agricultural Organization estimates that there are about 50,000 plants. About 30,000 plants have been documented out of around 17,810 plant species. Approximately 2200 species are categorized as medicinal plants, with an annual growth rate ranging from 7 to 15%. Global open research is built on these medicinal plants [2].

*Syzygium cumini* (S. cumini) was a plant belonging to the Mytraceae family that was also known by the names Jamun, jambul, jambolan, Indian blackberry, Java plum, and black plum. The *S. cumini* was also known by the synonyms Eugenia jambolana, Syzygium jambolana and *Eugenia cumini*. The *S. cumini* was a tropical evergreen tree with coarse discolored lower bark and white, gray stems that can reach heights of 25–30 meters (80–100 ft). The simple dark green, opposite, glossy, smooth, leathery-feeling leaves are blunt or narrow at the point of the leaf. They can also be elliptical or oblong-oval in shape. The size of the leaves is 2–8 centimeters wide and 5–15 centimeters long. There was one big 2 cm long seed in the fruit [3].

The tree only bears fruit once a year and the berries have a somewhat sweet and tart flavor [4]. The flower appears in March to April and the fruit develops in the months of May to July, approximately 32 days after the flower appears. The *S. cumini* fruit is often harvested in Asia between June and July, during the monsoon season, and the harvesting period lasts about thirty to forty days [5].

The Syzygium species can be found in tropical and sub-tropical regions across the world [6]. Typically, the Syzygium genus was grown in rainforests, including peat swamp forests, bamboo forests, monsoon-like forests, coastal forests and swamp forests [7].

The *Syzygium cumini* (*S. cumini*) was a huge evergreen tropical tree named Linn. (Syn. Eugenia, jambolan). It was a member of the Mytraceae family and was also known as Jamun, black plum or jambolan. The *S. cumini* was a promising fruit crop of the twenty-first century [8].

The *S. cumini* plant has been widely utilized in traditional medicine due to its abundance of bioactive chemicals present in all parts of the plant. The main bioactive components found in the edible portion are oxalic acid; tannins, gallic acid, malice acid, and oxalic acid are responsible for the therapeutic properties. These bioactive substances may help to prevent or reduce metabolic disorders and various kinds of diseases [9]. The *S. cumini* has a popular ingredient due to its high levels of vitamin C and anthocyanins [10]. Herbal medicines provide a promising choice to modern synthetic drugs as they show minimum or no side effects and are considered to be safe [11].

Secondary metabolites are natural chemicals derived from the *S. cumini* plant and are considered to be the most significant bioactive substances. Traditionally, a variety of diseases and illnesses have been treated with plant extracts. The extracts from the parts of *S. cumini* have been used to prevent and treat a number of illnesses and these days, their main uses were for their antidiabetic properties [12]. The *S. cumini* L. rapidly growing evergreen tropical tree of medium-to-large size is well known for their numerous applications in woodwork, remedies, fruit, food and pharmaceuticals, among other things. Almost every component of the plant was extremely important. The genus Syzygium was also utilized in culinary some species, like *S. aromaticum* are used for their unopened flower buds, which are used as herbs and are quite significant economically [13].

## 2.0 MATERIALS AND METHODS

### 2.1 Collection of Plant

Plant materials were collected from Charsadda Khyber

Pakhtunkhwa Pakistan.

2.2 Extraction and Isolation

The collected plant parts (leaves (A), stem (B) and root (C) of S. cumini were shade dried, cut into small fragments and grounded to a fine powder using an electric grinder. The powder materials were soaked in commercial-grade methanol for 15 days at room temperature with periodic shaking. After that, the material was filtered and the filtrate was concentrated at 40 oC by using a rotary evaporator.

The obtained extracts were suspended in distilled water and treated with a polar and non-polar solvent which yielded hexane, chloroform and methanolic extracts. Among chloroform fractions was assessed for column chromatography analysis using silica gel.

**2.3 Phytochemical Screening**

The phytochemical screening of the test samples were perform to qualitatively identify the alkaloids, tannins, saponins, flavonoids and anthocyanins present in medicinal plant according to standard procedure [14].

**2.2.1 Test for alkaloid**

After cooling 0.5 g of crude methanolic extract were heated for three minutes with 2% sulfuric acid ($H_2SO_4$) then filtered. Each extract received 2 mL of Dragendorff's reagent. The presence of an orange-red precipitate indicates the presence of alkaloids [15].

**2.2.2 Test for Flavonoid**

After dissolving 1 g of plant material in 5.0 mL of methanol, add 0.5 g of magnesium and a few drops of concentrated HCL to the mixture. The presence of flavonoids in the sample is indicated by the emergence of pink colors within 3 minutes [16].

**2.2.3 Test for Saponin**

By adding 2 g of the dried powdered samples was boiled in 20 mL of distilled water in a water bath and filtered. 10 mL of the filtrate was mixed with 5ml of distilled water and shaken vigorously for a stable froth. The formation of stable foam appears to indicate that saponins are present in the sample [17].

**2.2.4 Test for tannin**

By adding 0.5 g of the dried powdered samples was boiled in 20mL of water in a separate test tube and then filtered. A few drops of 0.1% of ferric chloride solution were added to each test tube separately. The presence of tannins is indicated by the development of a dark blue or greenish black color [18].

**2.2.5 Test for Anthocyanin**

The presence of anthocyanins has been demonstrated by adding 2 mL of the plant extract with 2 mL of 2 N HCl. The formation of a pink-red color of a pink-red color that turns purplish blue after addition of ammonia indicates the presence anthocyanins [19].

**2.4 Antibacterial Activity**

The crude methanolic extracts of S. cumini leaves (A), stem (B) and root (C) were screened for potential antibacterial activity against the test pathogens (P. aeruginosa, S. aureus, S. typhi and E. coli). Nutrient agar media was prepared, autoclaved and poured into sterilized petri plates. After incubation, the test organisms were inoculated into the sterile nutrient broth and incubated for 24 hrs at 37oC. In the laboratory, we used a sterile borer to make small wells (6 mm) in the petri plates. The stock solutions (3 mg/ml of DMSO) were transferred into the respective wells and the plates were incubated at 37°C for 24 hours. We used amoxicillin as a positive control and less than 1% DMSO as a negative control. The zones of inhibitions were recorded after 24 hrs of incubation at 37 ºC [20].

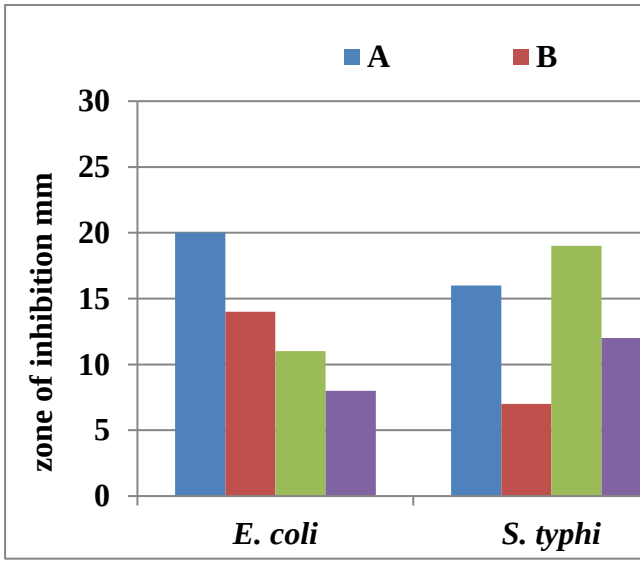
**2.7 Insecticidal Activity**

Insecticidal activity was performed by using contact toxicity assay against various insects *(R. Dominica, C. analis and T. castaneum*). On the first day, the (90 mm) petri plates were sterilized and we prepared papers cut to fit the size of the filter paper. The filter papers were placed within the petri plates and the stock solutions (200mg/ 3 mL) of the test samples were introduced using a micropipette. After complete evaporation of organic solvents from the petri plates on the second day, 16 healthy insects of each species were carefully selected and transferred to the labeled plates by using a clean brush. The plates containing samples and insects were incubated for 24 hours at

27°C in growth chamber with 50% relative humidity. In this study, we used the standard insecticidal drug permithrine as the positive control and the organic volatile solvent (methanol) was used as the negative control for the experiment. After incubation, the results were recorded by counting the number of survived insects in each plate [23].

**RESULTS AND DISCUSSION**

The crude methanolic extract *S. cumini* leaves (A), (B), (C) and (D) were screened for antibacterial activity against different test pathogens (*E. coli, S. typhi, S. aureus and P. aeruginosa*). The test sample A showed good activity against *E. coli* (20 mm), *S. typhi* (14 mm) and moderate against *S. aureus* (11 mm) while low antibacterial activity against *P. aeruginosa* (8mm), respectively. The test sample B showed good activity against *S. aureus* (19 mm), *E. coli* (16mm) and moderate activity against *P. aeruginosa* (12 mm), while low antibacterial activity against *S. typhi* (7 mm), respectively. The test sample C showed good antibacterial activity against *P. aeruginosa* (18 mm), *S. aureus* (15 mm) and moderate against *S. typhi* (12 mm), while low antibacterial activity against *E. coli* (9mm), respectively. The sample (D) showed significant activity against *E. coli* (25 mm) and *S. typhi* (22 mm), good antibacterial activity *S. aureus* (18 mm), *P. aeruginosa* (11 mm), respectively. In another study it was also reported that *the S. zeylanicum* root bark has been utilized in traditional medicine to treat infections caused by harmful bacteria. The bark and roots of *S. zeylanicum*, as well as to assess the antimicrobial activity of endophytic fungi and their secondary metabolites. The endophytic fungi from *S. zeylanicum* root bark are isolated and identified and their antibacterial activity against test pathogens was then evaluated [26].




Insecticidal Activity

The crude methanolic extract of S. cumini leaves (A), stem (B), root (C) and isolated compound (D) were determined against different pests: *Tribolium castaneum, Rhyzopertha Dominica and Callosbruchus analis.* The test sample A showed good activity against T. castaneum (58%) and moderate against R. dominica (30%) while low insecticidal activity against C. analis (19%) respectively. The test sample B showed good activity against R. dominica (55%) and moderate against C. analis (35%) while low insecticidal activity against T. castaneum (16%), respectively. The test sample C showed good activity against C. analis (60%) and moderate against T. castaneum (39%) while low insecticidal activity against R. dominica (18%), respectively. The isolated compound D showed significant activity against T. castaneum (92%), good showed against R. dominica (66%) while low activity against C. analis (25%), respectively. Our results also supported by another study, that the chemical component present in S. lineare leaves has insecticidal activities against (spodoptera litura, spodoptera litura Fabricius, and Danaus plexippus). According to this study, the insecticidal substance 2,3-diacetoxy-2-benzyl-4,4,6,6-tetramethyl-1,3-cyclohexanedione from S. lineare leaves has been successfully isolated and described. Additionally, the study proved that it had strong insecticidal properties [27].

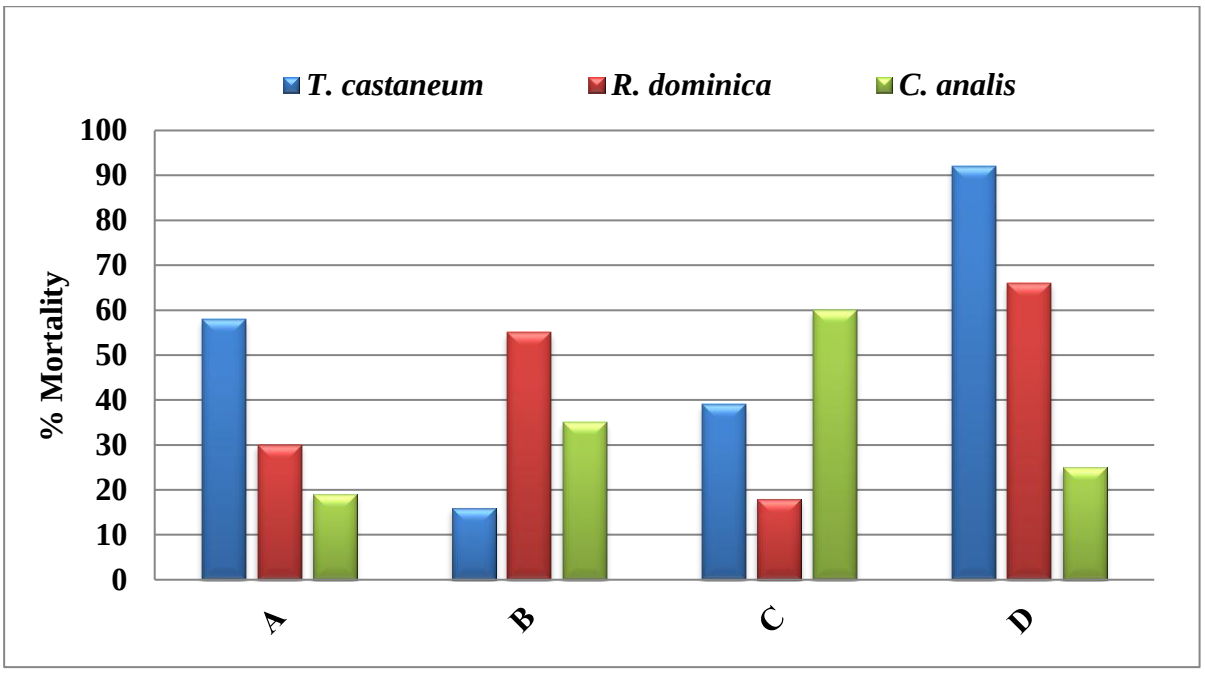



Figure 2. Insecticidal activity of crude methanolic extract of *S. cumini* STD (*Permithrine*)

## 4.0 DISCUSSION

The antibacterial activity of the was evaluated against six bacterial strains, including *Bacillus subtilis*, *Staphylococcus aureus*, *Enterobacter*, *Pseudomonas aeruginosa*, *Escherichia coli*, and *Salmonella Typhi*. The results demonstrated that significantly inhibited bacterial growth, with the highest inhibition observed for *S. aureus* and *K. pneumoniae* which is consistent with findings in other. These exhibited effective bactericidal action, suggesting their potential application in combating multidrug-resistant (MDR) strains, which are a major global health concern [28].

The antibacterial and antifungal effects depend not only on their concentration but also on their ability to penetrate bacterial cell membranes and disrupt cellular functions. enhancing their antimicrobial efficacy.

## 5.0 CONCLUSION

This study demonstrates the successful use of *S. cumini crude* extract, providing an eco-friendly, cost-effective, and scalable method for medicinal production. The presence of key phytochemicals in the extract played a crucial role in the reduction and stabilization. The leaves and roots extract provide significant antibacterial activity against pathogenic bacterial strains, including *S. aureus* and *K. pneumoniae*, highlighting their potential for medical applications. Furthermore, the nanoparticles demonstrated antioxidant properties, supporting their broader therapeutic potential. This approach not only provides an effective method for producing therapeutic products but also contributes to the development of sustainable products for biomedical and industrial use [29].

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## CONFLICT OF INTEREST

Declare conflicts of interest or state “The authors declare no conflict of interest

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