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## Research Article

# Exploring the Bioactive Potential of *Desmodium elegans*: Phytochemical and Biological Insights

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## ABSTRACT

The biological potential of *Desmodium elegans* (*D. elegans*), roots and leaves were evaluated through various in vitro assays. Phytochemical screening revealed the presence of several bioactive secondary metabolites, including alkaloids, anthraquinones, saponins, phenols, and tannins. The antimicrobial, phytotoxic, antioxidant, and insecticidal activities of the plant extracts were assessed using the direct contact method, agar well diffusion method, and 2,2-diphenyl-1-picrylhydrazyl (DPPH) radical scavenging assay. The crude root extract demonstrated significant antibacterial activity, inhibiting *Escherichia coli* (11 mm), *Salmonella Typhi* (6 mm), and *Pseudomonas aeruginosa* (11 mm). In contrast, the leaf extract showed stronger inhibition against *Klebsiella pneumoniae* (14 mm) and moderate activity against *E. coli* (13 mm), *S. Typhi* (14 mm), and *P. aeruginosa* (14 mm). The methanolic extract of the root exhibited minimal antifungal activity, with only 5% inhibition against *Aspergillus flavus* and 10% against *Trichoderma harzianum*. The antioxidant potential of *Desmodium elegans* (root and leaves) was also noteworthy, with scavenging activities of 42% and 46% at 60 ppm, and 51% and 57% at 80 ppm, respectively. Phytotoxicity assays revealed the root extract exhibited 25% and 6.25% activity, while the leaf extract demonstrated 31.25% and 25% activity at concentrations of 1000 µg/ml and 100 µg/ml. Insecticidal assays revealed positive results against *Tribolium castaneum*, *Rhyzopertha dominica* (20%), and *Callosobruchus analis* (60%) at higher concentrations. These findings suggest that *Desmodium elegans* possesses significant bioactive potential and could serve as a valuable source of natural products for antimicrobial, antioxidant, and pest control applications.

**Keywords:** *Desmodium elegans*; Antimicrobial Activity; Antioxidant; Insecticidal Activity; Phytotoxicity.



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## INTRODUCTION

Plants are an invaluable gift to humanity, contributing to the sustenance of life on Earth in multiple ways. Often referred to as the "lungs of the Earth," they play a crucial role in the production of oxygen through photosynthesis while simultaneously absorbing carbon dioxide, which is vital for life on our planet (Soetan et al., 2010; Tomlinson and Akerele, 2015). Beyond these essential ecological functions, plants offer a wide range of benefits, including food, shelter, raw materials, and emotional support. For centuries, they have been integral to human survival, providing essential nutrients and medicinal resources that promote health and well-being across diverse cultures. Their contributions in maintaining ecological balance and enhancing biodiversity are irreplaceable. Moreover, plants serve as the foundation for numerous essential goods, ranging from food to pharmaceuticals, which help sustain human and animal life across the globe (Abdur Rauf et al., 2012).

Among the most significant roles of plants is their use in traditional and modern medicine. Medicinal plants, known for their therapeutic properties, have been employed for centuries to treat various ailments. From early civilizations in Africa to Indigenous peoples of the Americas, plants have formed the basis for numerous healing practices, serving as natural remedies for a wide array of diseases (Abdur Rauf et al., 2012; Rauf et al., 2012). As societies evolved, so did the understanding and utilization of plant-based remedies. Ethnobotany, the study of the relationship between plants and humans, seeks to preserve and document this knowledge. Today, modern scientific research and technological advancements have allowed us to investigate the chemical properties of plants at a molecular level, enabling us to extract and analyze the bioactive compounds responsible for their medicinal effects. These bioactive compounds such as alkaloids, flavonoids, terpenes, and polyphenols have demonstrated potent antimicrobial, anti-inflammatory, antioxidant, and anticancer activities (Davis and Choisy, 2024; Guy-Rodolphe N'cho et al., 2025; Tomlinson and Akerele, 2015).

One plant that has garnered significant attention for its medicinal potential is *D. elegans*, a member of the Fabaceae family. This plant has been widely recognized for its therapeutic properties and is increasingly being explored for its possible applications in modern medicine. The growing interest in *D. elegans* stems from its affordability, widespread availability, and low toxicity, making it a promising candidate for the development of new, cost-effective treatments, particularly in regions where access to synthetic drugs is limited (Balakumar et al., 2011; Cueva-Chamba et al., 2023). The plant is being studied for its antibacterial, anti-inflammatory, and antioxidant effects, which could lead to the development of novel therapeutic agents with fewer side effects than conventional pharmaceuticals (Balakumar et al., 2011; Davis and Choisy, 2024).

The *Desmodium* genus, which includes *D. elegans*, is rich in bioactive compounds, particularly alkaloids. Alkaloids are nitrogen-containing compounds known for their potent therapeutic effects, including antimicrobial, anti-inflammatory, and analgesic properties (Balakumar et al., 2011; Zhi KangKang et al., 2014). These compounds have been extensively studied for their potential in drug development, as they can offer new ways to treat diseases with fewer adverse effects compared to synthetic drugs. Additionally, the *Desmodium* species has been found to synthesize other bioactive secondary metabolites, including flavonoids and terpenes, which further increase its therapeutic potential (Pant and Thakur, 2021). Research into the alkaloids found in *D. elegans* has provided new insights into their medicinal properties, making the plant an exciting subject for pharmacological research and the development of new drugs (Zhao et al., 2019).

*Desmodium elegans* is an erect, woody deciduous shrub that grows to a height of 1.8 to 2.5 meters. The plant is characterized by its trifoliate leaves and purple flowers that bloom from August to September. Its fruits are small, hanging from the stem, and the seeds are dispersed from September to December. The plant is widely distributed across South Asia, including Kashmir, India, Nepal, and Bhutan, where it thrives in a variety of soil types, from sandy to loamy soils. In Pakistan, *D. elegans* is commonly found in the Gallyat regions of Khyber Pakhtunkhwa, where its ability to adapt to various environmental conditions allows it to flourish (Ahmad Khan and Ahmad, 2019; Tomlinson and Akerele, 2015). The plant's ecological versatility and resilience make it a valuable resource in both natural and agricultural systems.

The medicinal uses of *Desmodium elegans* are diverse, with various parts of the plant including the roots, leaves, flowers, and seeds being utilized in traditional medicine. The roots are known for their diuretic properties, which help in increasing urine production and promoting digestion. In certain regions, the powdered leaves are used to treat wounds and injuries, showcasing the plant's natural antiseptic and healing qualities. Additionally, the plant has been used to treat conditions such as fever, stomach problems, and lung diseases, highlighting its broad therapeutic spectrum (Joshi et al., 2023; Ma et al., 2011). The rich chemical composition of *D. elegans*, including the presence of alkaloids, flavonoids, and other secondary metabolites, further supports its value as a source of natural remedies (George et al., 2025).

Recent research into the phytochemical properties of *D. elegans* has revealed its potential as a source of antimicrobial agents. Alkaloids isolated from the plant have demonstrated significant antibacterial activity against a variety of pathogens, including *Escherichia coli*, *Salmonella typhi*, and *Pseudomonas aeruginosa* (Gitu, 2013). Furthermore, the plant's antioxidant properties, as determined by radical scavenging assays, suggest that it may offer protection against oxidative stress, which is linked to a range of chronic diseases, including cancer and cardiovascular disorders. These findings underscore the promise of *D. elegans* as a therapeutic agent and provide a scientific basis for its traditional use in treating infections and inflammatory conditions.

Despite its historical significance in traditional medicine, research on the full range of medicinal properties of *D.*

*elegans* remains limited. Much of the existing literature has focused on the plant's bark and stems, with less attention given to the roots and leaves. These parts of the plant have been less explored in terms of their bioactive compounds, which represent a promising avenue for further investigation (Mungwari et al., 2025). Exploring the roots and leaves of *D. elegans* could uncover new secondary metabolites with potential therapeutic applications, addressing a critical gap in the current body of research. This study aims to investigate the biological and phytochemical properties of the roots and leaves of *Desmodium elegans*, with a focus on their antimicrobial, antioxidant, and anti-inflammatory effects.

## MATERIALS AND METHODS

### General Experimental Conditions

All chemical, biological, and instrumental analyses were conducted at the Institute of Biotechnology and Microbiology at Bacha Khan University Charsadda.

### Collection of Plant

*Desmodium elegans* was collected from the Gallyat region of Khyber Pakhtunkhwa, Pakistan.

### Extract Preparation

The collected *D. elegans* plant was shade-dried, chopped into small pieces, and ground into a fine powder using an electric grinder. The powdered material was then soaked in commercial-grade methanol for 15 days, with occasional shaking. After the soaking period, the material was filtered, and the filtrate was concentrated under vacuum at 40°C using a rotary evaporator.

### Phytochemical Screening

Phytochemical screening of the *Desmodium elegans* extracts was conducted to detect the presence of bioactive compounds such as alkaloids, anthraquinones, saponins, tannins, and phenols, following standard methods (Guy-Rodolphe N'cho et al., 2025; Joshi et al., 2023). Various tests were performed to identify these compounds. To detect tannins, 2 mL of 5% ferric chloride solution was added to 1 mL of plant extract, and the appearance of a greenish-black or dark blue color confirmed the presence of tannins. For the detection of saponins, 5–10 mL of distilled water was added to 1 mL of the plant extract, which was then shaken for 15 minutes in a graduated cylinder. The formation of a 1 cm foam layer indicated the presence of saponins. To test for alkaloids, 2 mL of plant extract was mixed with 2 mL of concentrated hydrochloric acid, followed by the addition of 3 drops of Mayer's reagent. A green color or the formation of a white precipitate confirmed the presence of alkaloids. For anthraquinones, 0.5 g of plant extract was boiled with 10% HCl for several minutes, and after cooling, equal volumes of chloroform and 10% ammonia were added. The appearance of a rose-pink color indicated the presence of anthraquinones. Lastly, for phenols, 1 mL of the plant extract was mixed with 2 mL of distilled water and 5 drops of 10% ferric chloride solution. The formation of a blue or green color indicated the presence of phenols.

### Antifungal Activity

Antifungal activity was assessed using the agar tube slant method (Mazher et al., 2017). Stock solutions of the extract (24 mg/mL) were prepared by dissolving the extract in dimethyl sulfoxide (DMSO). The nutrient media, consisting of 66 g of Sabouraud Dextrose Agar (SDA) dissolved in 1000 mL of distilled water, was mixed thoroughly and heated. After autoclaving the media at 121°C for 15 minutes, 4 mL of the media was added to screw-cap tubes, followed by the addition of the stock solution. These tubes were allowed to solidify in a slanted position at room temperature. Each tube was inoculated with fungal species such as *A. niger*, *A. flavus*, *T. harzianum*, and *R. stolonifer*. Inhibition of fungal growth was recorded after 7 days of incubation (Akomah-Abadaike and Didia, 2024).

### Antibacterial Activity

Antibacterial activity was evaluated by the agar well diffusion method (Din et al., 2024). Nutrient agar plates were inoculated with fresh bacterial cultures, including *E. coli*, *S. typhi*, *S. aureus*, *P. aeruginosa*, and *K. pneumoniae*, using a sterile cotton swab to ensure efficient growth. Wells were made in each plate using a cork borer, and 0.5 mL of various extracts were added to the wells. The zone of inhibition was measured by calculating the diameter of the inhibition zone, including the well diameter. The results were averaged from three replicates in different fixed directions (Din et al., 2024).

### Antioxidant Activity

The antioxidant activity of the crude extract of *D. elegans* (root and leaves) was measured using the DPPH (2,2-diphenyl-1-picrylhydrazyl) radical scavenging assay, as reported in previous studies (Fouda et al., 2023). Stock

solutions of 25 mg/50 mL were prepared, and different concentrations (20, 40, 60, 80, and 100 ppm) were made. In each solution, 1 mL of 0.001M DPPH was added to 2 mL of the crude extract. The absorbance was measured at 517 nm after 30 minutes using a SP-3000 PLUS Spectrophotometer (Optima, Japan). Percent radical scavenging activity (RSA) was calculated using the formula:

$$\text{Percent RSA} = ((\text{Control absorbance} - \text{Sample absorbance}) / \text{Control absorbance}) * 100$$

#### Phytotoxic Activity

Phytotoxic activity was assessed by screening the test samples for their effects on the growth of *Lemna minor* plants (Coronado-Posada et al., 2013). Stock solutions of the test samples (10, 100, 1000 µg/mL) were prepared and poured into flasks containing sixteen healthy *L. minor* plants. The flasks were incubated for 7 days, and the results were recorded based on the growth inhibition observed (Irfan et al., 2024).

#### Insecticidal Activity

The insecticidal activity of *D. elegans* was evaluated using a contact toxicity assay (Akomah-Abadaike and Didia, 2024). A filter paper was placed in a petri dish, and stock solutions of the test samples were applied using a micropipette. After 24 hours of incubation at 27±1°C, 16 healthy insects from selected species (*T. castaneum*, *R. dominica*, *C. analis*) were transferred into each petri dish. The number of surviving insects was recorded to evaluate the insecticidal efficacy of the extracts (Lu et al., 2020).

#### Statistical analysis

A one-way analysis of variance (ANOVA) was conducted using SPSS 18.0 to assess significant differences among the experimental groups. Post hoc comparisons were performed using Duncan's multiple range test to identify specific group differences at a 5% significance level ( $P < 0.05$ ). This test is suitable for comparing multiple group means while controlling for Type I error, ensuring reliable pairwise comparisons when ANOVA indicates significant overall variation (Din et al., 2023).

## RESULTS AND DISCUSSION

### Phytochemical Investigations

Phytochemical screening of the crude extracts of *Desmodium elegans* (roots and leaves) was carried out to identify the presence of various bioactive compounds, including alkaloids, tannins, saponins, anthraquinones, and phenols. As shown in Table 1, all tested compounds were found to be present in both the root and leaf extracts of *D. elegans*. Alkaloids were detected through the formation of a green color after the addition of Mayer's reagent, and the presence of saponins was confirmed by the formation of a 1 cm foam layer upon shaking the extract with distilled water. The test for tannins was positive, as indicated by the greenish-black or dark blue color formed when ferric chloride was added. Anthraquinones were identified by the appearance of a rose-pink color, and phenols were detected by the blue or green color formed after adding ferric chloride (Ali et al., 2018; Coronado-Posada et al., 2013). These findings suggest that *D. elegans* contains a range of bioactive compounds that could contribute to its medicinal properties. The presence of alkaloids, in particular, indicates potential for antimicrobial and anti-inflammatory activities, which aligns with the plant's traditional uses in folk medicine.

Table 1. phytochemical screening of crude extract of *D. elegans* (Root, Leaves)

S. No	Compounds	<i>D. elegans</i>	
		Root	Leaves
01	Alkaloids	+	+
02	Tannins	+	+
03	Saponins	+	+
04	Anthraquinones	+	+
05	Phenols	+	+

### Antifungal Activity

The antifungal activity of the crude extracts of *D. elegans* was tested against *A. niger*, *A. flavus*, *T. harzianum*, and *R. stolonifer*. As depicted in Figure 1, both the root and leaf extracts of *D. elegans* showed no significant activity against *A. niger* and *R. stolonifer*. However, the root extract exhibited a low inhibition of 5% against *A. flavus*, while the leaf extract showed a slightly higher inhibition at 9%. Additionally, the root extract showed 10% inhibition against *T. harzianum*, while the leaf extract displayed no activity. These results suggest that *D. elegans* has limited antifungal

potential. The statistical analysis of these results, presented in terms of p-values, confirmed that the differences in inhibition between the root and leaf extracts were not significant ( $p > 0.05$ ). Although the plant is traditionally used in various cultures for its medicinal benefits, the scientific evaluation reveals that its antifungal efficacy is not as significant as initially expected (Kaur et al., 2021). Historically, many plant oils and extracts have been used as topical antiseptics or for their antimicrobial properties, and the results of this study further emphasize the importance of scientifically validating traditional medicinal uses (Gitu, 2013). Error bars representing the mean  $\pm$  standard deviation are shown in the figures, indicating the variability of the data from three independent experiments.

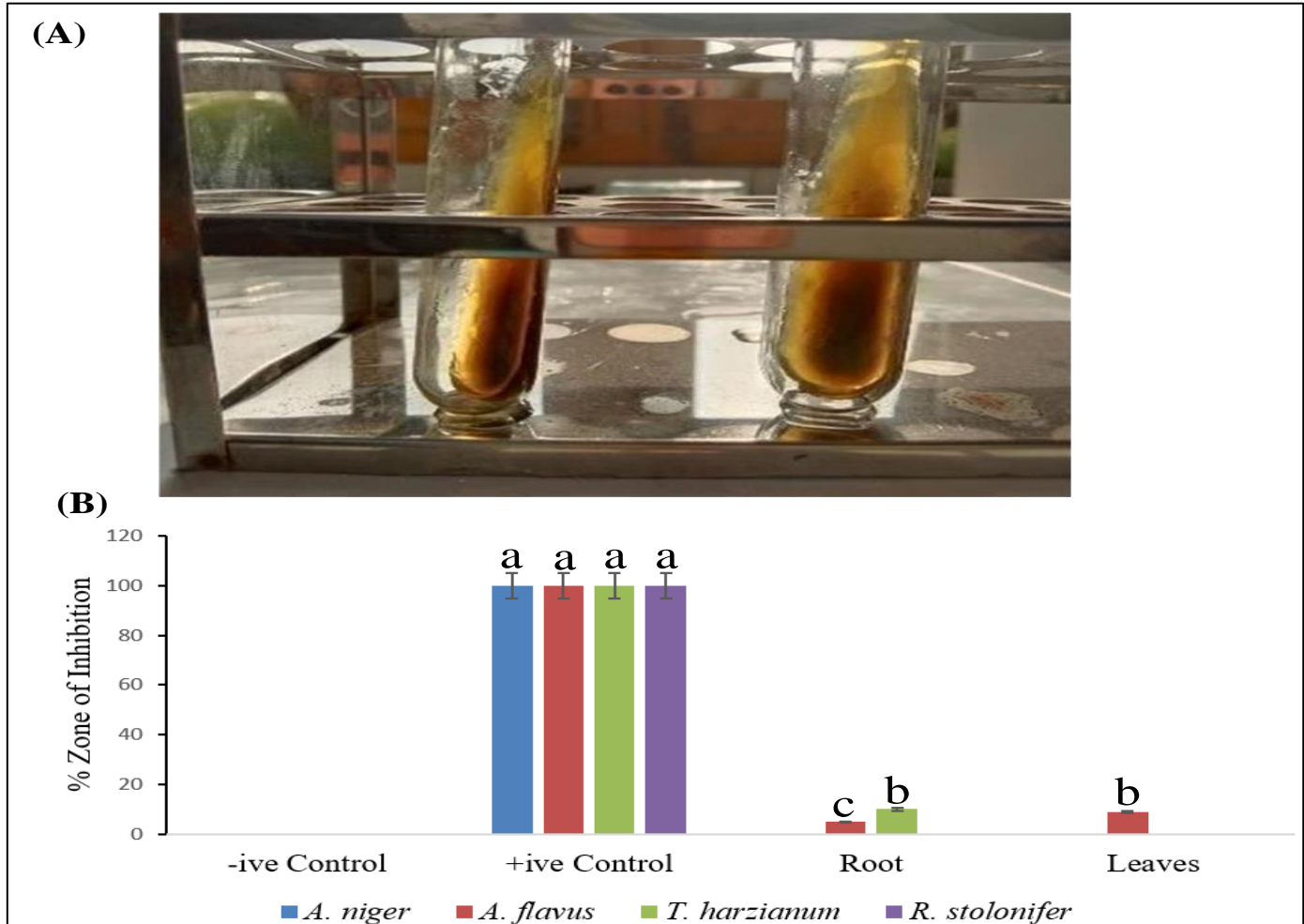


Figure 1. (A): antifungal activity of the crude extracts of *D. elegans* in slant (B): Diameter of the inhibition zones. Error bars presented are the mean  $\pm$  standard deviation of triplicates of three independent experiments.

Note: Different letters indicate a significant difference ( $p = 0.05$ ). Means with the same letters are not significantly different at  $p < 0.05$ .

### Antibacterial Activity

The antibacterial activity of *Desmodium elegans* leaf extract was assessed against several common bacterial pathogens, including *E. coli*, *S. typhi*, *S. aureus*, *P. aeruginosa*, and *K. pneumoniae*. The results, presented in Figure 2, show that the leaf extract exhibited strong activity against *K. pneumoniae* (66.66%) and moderate activity against *E. coli* (48.14%), *S. typhi* (51.85%), and *P. aeruginosa* (51.85%) compared to the positive control, streptomycin. Additionally, the leaf extract demonstrated lower activity against *S. aureus* (38.46%). These findings suggest that the leaf extract of *D. elegans* possesses significant antibacterial potential. Statistical analysis of the results, including p-values and error bars, confirmed that the observed antibacterial activities were statistically significant ( $p < 0.05$ ). The variability in the data is represented by the mean  $\pm$  standard deviation from three independent experiments. These results are particularly relevant given the increasing issue of multidrug resistance in bacterial pathogens, such as *S. aureus*, which has become resistant to several common antibiotics (Kong et al., 2014; Zarroug et al., 2023). This highlights the growing importance of plant-derived antibacterial agents as alternatives to synthetic drugs, especially in regions where antibiotic resistance is a major concern.

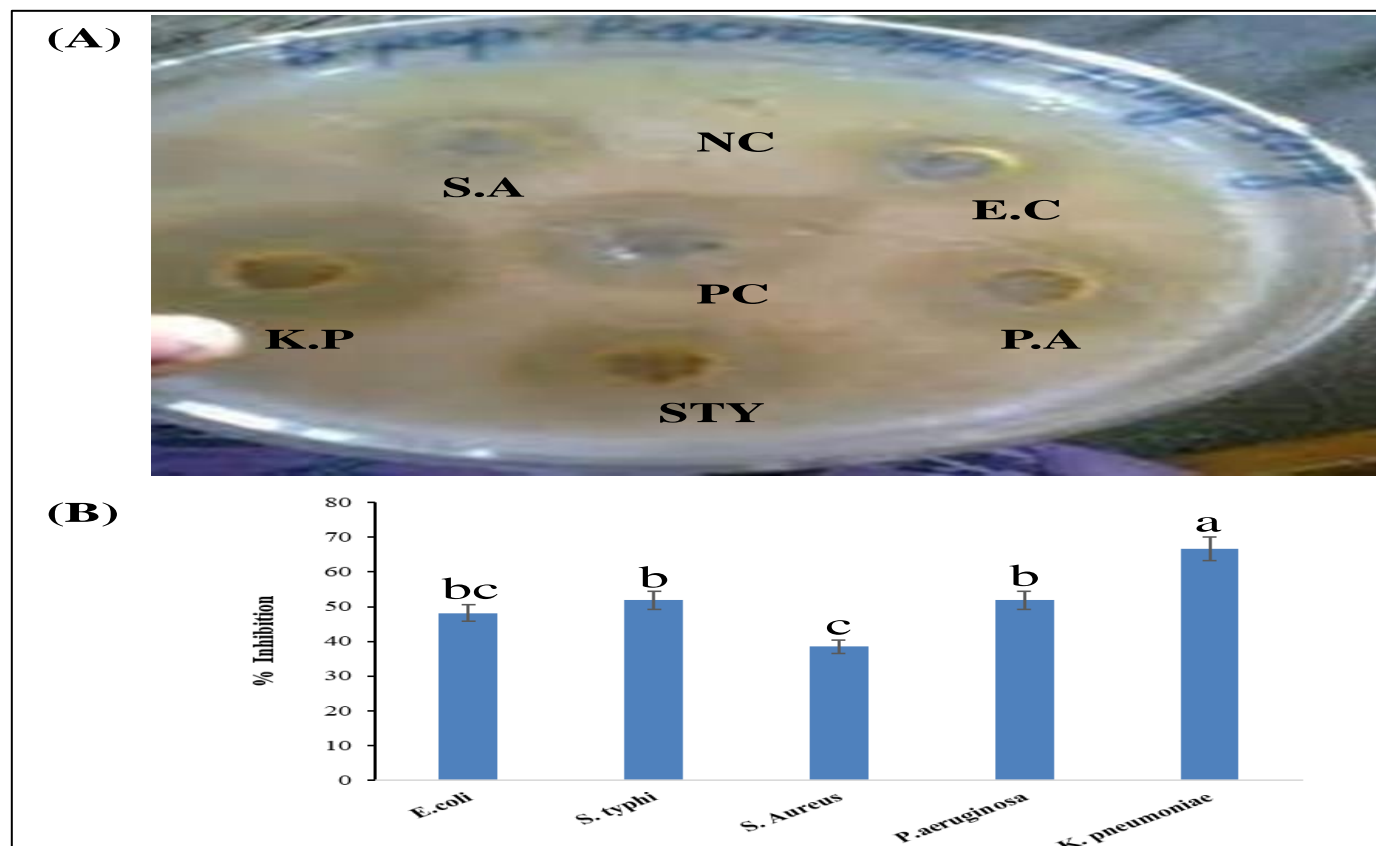


Figure 2. (A) Antibacterial activity of the crude extract from *D. elegans* leaves. (B) Diameter of the inhibition zones. Error bars represent the mean  $\pm$  standard deviation from triplicates of three independent experiments. Different letters indicate a significant difference ( $p = 0.05$ ). Means with the same letters are not significantly different at  $p < 0.05$ .

Note: The abbreviations used in the figure are as follows: PC stands for Positive Control (streptomycin), NC for Negative Control, KP for *Klebsiella pneumoniae*, EC for *Escherichia coli*, STY for *Salmonella typhi*, SA for *Staphylococcus aureus*, and PA for *Pseudomonas aeruginosa*.

### Antioxidant Activity

The antioxidant activity of the crude extracts of *D. elegans* was assessed using the DPPH free radical scavenging assay. As shown in Figure 3, both root and leaf extracts demonstrated significant antioxidant activity at varying concentrations. At 40 ppm, the leaf extract exhibited 22% radical scavenging activity, while the root extract showed 17%. At higher concentrations (60 ppm), the leaf extract showed 46%, and the root extract showed 42%. The highest concentration tested (100 ppm) resulted in 74.5% scavenging activity for the leaf extract and 79.4% for the root extract. These results suggest that *D. elegans* possesses moderate to high antioxidant activity, with the leaf extract exhibiting slightly better activity than the root extract. Statistical analysis, including p-values and error bars, confirmed that the differences in antioxidant activity between the root and leaf extracts were statistically significant ( $p < 0.05$ ). The variability of the results is represented by the mean  $\pm$  standard deviation from three independent experiments.

The higher antioxidant activity of the leaf extract is likely attributed to the presence of flavonoids and phenols, which are known to have potent antioxidant properties (Ayuda-Durán et al., 2020; Zhang et al., 2016). The ability of *D. elegans* to scavenge free radicals supports its traditional use as an antioxidant agent and indicates its potential for use in the prevention of oxidative stress-related diseases.

### Phytotoxic Activity

The phytotoxic activity of *D. elegans* crude extracts was evaluated using the Lemna minor assay, a model system used to detect plant growth regulation. The results, shown in Tables 2 and 3, indicated that the root extract exhibited 25% growth inhibition at 1000  $\mu\text{g}/\text{mL}$  and 6.25% at 100  $\mu\text{g}/\text{mL}$ , while the leaf extract showed 31.25% and 25% inhibition at the same concentrations, respectively. No phytotoxic activity was observed at a concentration of 10  $\mu\text{g}/\text{mL}$ . Statistical analysis, including p-values and error bars, confirmed that the differences in phytotoxic activity between the root and leaf extracts were statistically significant ( $p < 0.05$ ), with the leaf extract showing a stronger effect at the higher concentration. The variability of the results is represented by the mean  $\pm$  standard deviation from three independent experiments.

These findings suggest that *D. elegans* extracts, especially the leaf extract, have potential as natural herbicides. Herbicides derived from plants are often considered environmentally friendly alternatives to synthetic herbicides, which can have harmful ecological effects. *L. minor*, a small aquatic monocot, is highly sensitive to bioactive compounds, making it an ideal model for screening phytotoxic compounds (Coronado-Posada et al., 2013; Guy-Rodolphe N'cho et al., 2025).

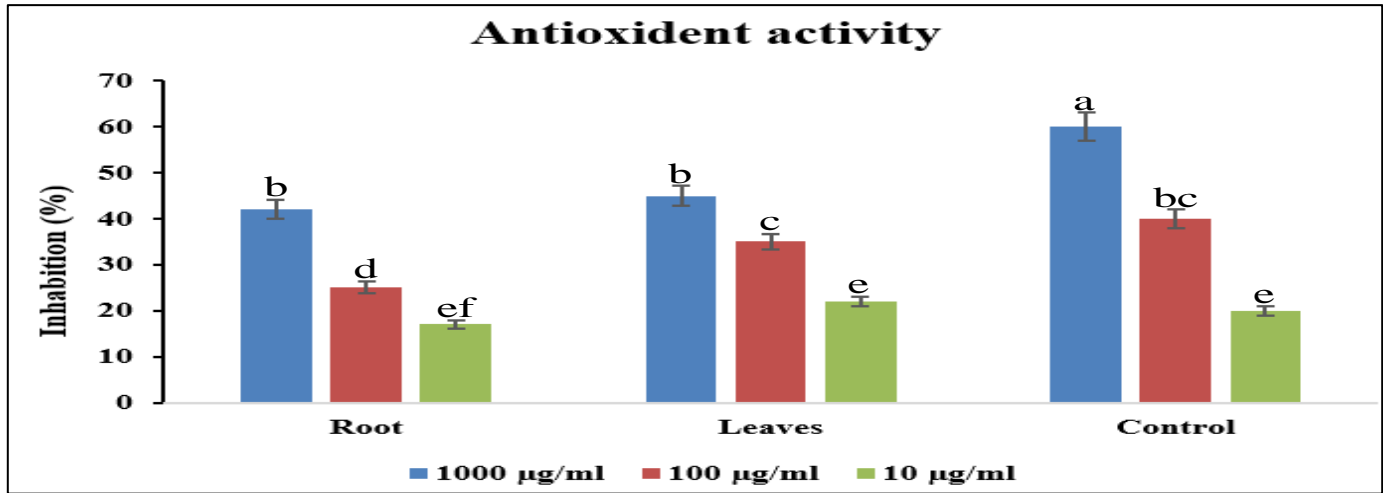


Figure 3. Antioxidant activity of crude extract of *D. elegans* (Root, Leaves)  
 Note: Different letters indicate a significant difference ( $p = 0.05$ ). Means with the same letters are not significantly different at  $p < 0.05$ .

Table 2. Phytotoxic activity of crude extract of *D. elegans* (Root, Leaves).

Name of plant	Concentration of sample (µg/ml)	No. of fronds survived			Conc standard drug * (µg/ml)
		Root	leaves	Control	
Lemna minor	1000	12	11	16	0.015
	100	15	12	16	
	10	16	16	16	

Table 3. Percent reduction in growth regulation of the *L. minor*.

Concentration of sample (µgm/ml)	Percent growth regulation	
	Root	Leaves
1000	25	31.25
100	6.25	25
10	0	0

### Insecticidal Activity

The insecticidal activity of the crude extracts was evaluated on *T. castaneum*, *R. dominica*, and *C. analis*. As shown in Table 7 and Figure 4, the root extract showed no activity against *T. castaneum* or *R. dominica*, while the leaf extract exhibited a 20% mortality rate against *T. castaneum* and 60% mortality against *C. analis*. The leaf extract was inactive against *R. dominica*.

Statistical analysis, including p-values and error bars, confirmed that the differences in mortality rates were statistically significant ( $p < 0.05$ ), with the leaf extract showing stronger activity against *C. analis* compared to *T. castaneum*. The variability of the results is represented by the mean  $\pm$  standard deviation from three independent experiments.

These results suggest that *D. elegans* leaf extract has potential as an insecticidal agent, particularly against *C. analis*. The environmental impact of synthetic insecticides is a growing concern, and plant-based insecticides offer a safer, more sustainable alternative (Ahmad et al., 2019; Mostafa et al., 2012).

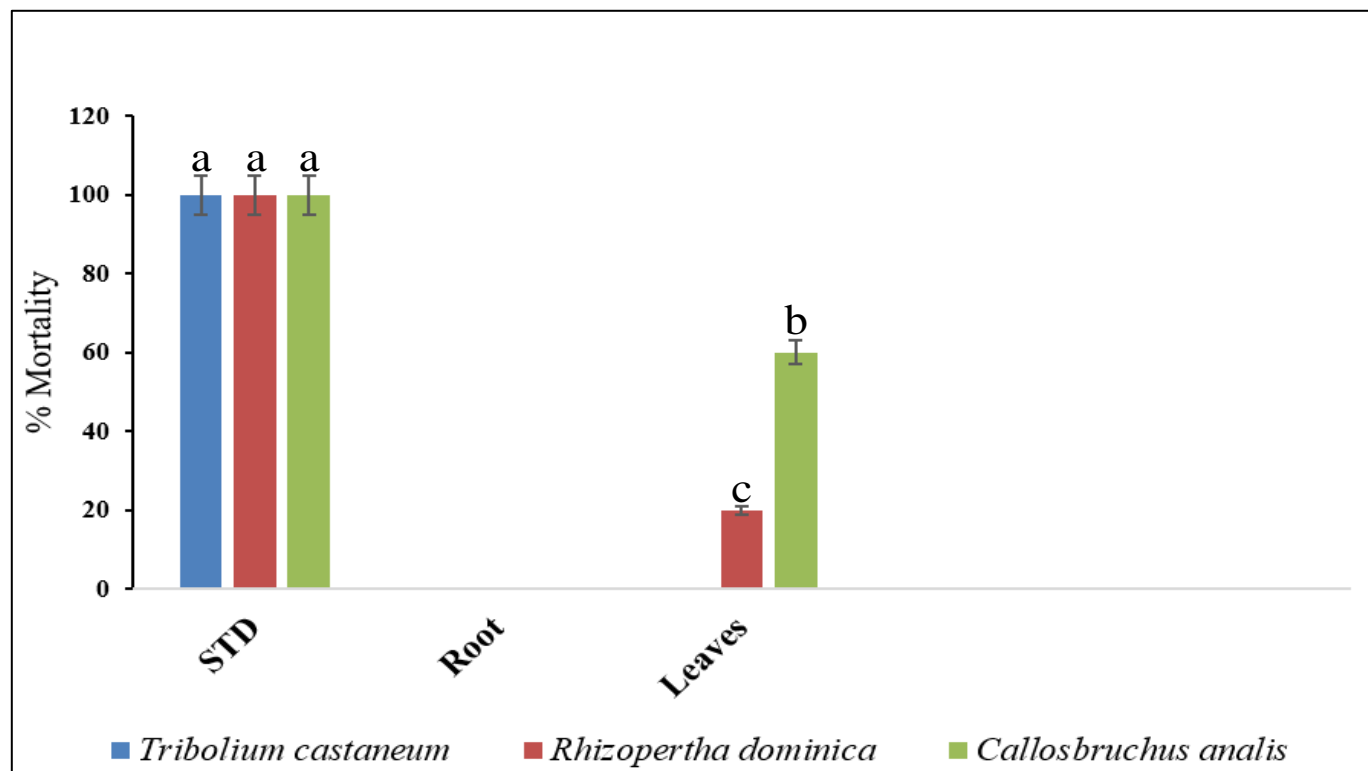


Figure 4. Insecticidal activity *D. elegans* (Root, Leaves). different letters indicate a significant difference ( $p = 0.05$ ). Means with the same letters are not significantly different at  $p < 0.05$ .

## CONCLUSION

The crude extract of *D. elegans* (root and leaves) demonstrates significant biological and phytochemical activity. Phytochemical screening revealed the presence of bioactive compounds such as alkaloids, anthraquinones, saponins, phenols, and tannins, which likely contribute to its moderate antimicrobial activity against pathogens like *E. coli*, *S. typhi*, *S. aureus*, *P. aeruginosa*, and *K. pneumoniae*. The leaf extract showed notable antioxidant properties, suggesting its potential as a natural antioxidant source. Additionally, the leaf extract exhibited stronger phytotoxic effects than the root, indicating its potential for plant growth regulation. The insecticidal activity of the extract, particularly at higher concentrations, suggests its application as a natural insecticide. Overall, *D. elegans* (root and leaves) shows considerable bioactivity, and further research could optimize its use in medicinal, agricultural, and biotechnological applications.

## AUTHOR CONTRIBUTIONS

J.N. and S.N. conceptualized and designed the research. Z.U.D., I.U.D., and I.U.D. were responsible for data collection. Z.U.D. and I.Z. conducted the data analysis. Z.U.D. and I.Z. also contributed to the writing of the manuscript. Z.U.D., M.A., and I.U.D. reviewed and edited the manuscript, while also performing software analysis and validation. All authors have read and approved the final manuscript.

## COMPETING OF INTEREST

The authors declare no competing interests.

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