



Antibacterial Activity of Sidr (*Ziziphus spina-christi*) Plant Extracts against Urinary Pathogens

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Abstract

Background: Urinary tract infections (UTIs) are common bacterial infections in children, with *Escherichia coli* being a primary causative agent. Traditional remedies like the Sidr plant have gained popularity due to their therapeutic properties, including antibacterial effects. **Methods:** Sidr leaves were collected, dried, and powdered. A cold aqueous extract was prepared by dissolving the powder in distilled water, and an alcoholic extract was made using ethyl alcohol. Chemical analysis was conducted to identify active ingredients. The antibacterial activity of both extracts was evaluated against bacterial isolates using agar well diffusion method. **Results and Discussion:** Chemical analysis revealed the presence of alkaloids, phenols, terpenoids, and other compounds in the Sidr plant. Both aqueous and alcoholic extracts showed antibacterial activity against all tested bacterial isolates. Increasing concentrations of the extracts correlated with increased inhibition of bacterial growth, with the alcoholic extract exhibiting a maximal inhibitory effect at 200 mg/ml. **Conclusion:** The antibacterial activity of Sidr plant extracts is attributed to their impact on bacterial cell activity and membrane permeability, likely due to the presence of phenolic compounds. These findings support

the potential use of Sidr plant extracts as alternative or adjunct therapies for UTIs.

Keywords: Zizyphusspin-csiti, Urinary tract infections, Sidr plant, Antibacterial activity, Herbal remedies, Microbial susceptibility.

Introduction

Infections of the urinary system caused by bacteria are among the most prevalent diseases in children (Bien et al., 2012). Second only to these in prevalence are respiratory tract infections (Shah et al., 2019). The majority of urinary tract infections (UTIs) are caused by enteric bacteria from the intestines, with *Escherichia coli* being the primary culprit. These infections typically occur when Gram-negative bacteria from the gastrointestinal tract invade the urinary system (Bhatt et al., 2012). Uropathogenic *Escherichia coli* (UPEC) is the most prevalent, responsible for 80-85% of UTIs (Nicolle, 2008).

While antibiotics are commonly used to treat bacterial infections, there has been a growing interest in natural remedies involving various herbs, which offer an alternative to synthetic drugs. One such significant plant is the Sidr (*Ziziphus spina-christi*), known for its therapeutic properties. The Sidr plant is utilized for treating a wide range of ailments, including headaches, fever, swollen and painful joints, dandruff, malignant tumors, malaria, and immunodeficiency diseases. Additionally, its leaves are known to purify and strengthen the intestines and skin and alleviate stomach pain (Nickel et al., 1998).

Sidr plant extracts have demonstrated high effectiveness against the growth of Gram-positive and Gram-negative bacteria and various fungi such as *Candida* species. This antimicrobial activity extends to pathogens like staphylococci and streptococci (Rushton, 1997).

Significance | Sidr plant extracts showed promising antibacterial activity against urinary pathogens as potential alternative of UTI treatments.

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The onset and progression of UTIs depend on complex interactions between the pathogen and the host, including the bacteria's virulence and ability to adhere to host tissues (Mahapatra and Heffner, 2019). Infections can spread to the urinary tract, urethra, kidneys, and bladder (DeVinney and Pitout, 2017; Kumar et al., 2014; Chaudhari et al., 2016). Severe cases may lead to conditions such as proteinuria, febrile UTIs, hypertension, chronic kidney disease, and renal scarring (Li et al., 2017).

Research has shown that the alcoholic extract of Sidr leaves is particularly effective against bacterial growth (Al-Abed, 2008). This study aims to explore the potential of Sidr plant extracts, using both cold water and ethyl alcohol, as a natural remedy for bacterial infections. By understanding the antibacterial effects of these extracts, we can evaluate their potential in treating various illnesses caused by these microorganisms.

Materials and Methods

Collecting Plant Samples

Sidr leaves were obtained from a local farm. After thoroughly cleaning the leaves with tap water to remove any dust, they were dried in a shaded area. Once dried, the leaves were ground into a fine powder. This leaf powder was then stored in dry, clean nylon bags and refrigerated for use in microbiological research.

Preparing the Chilled Aqueous Extract

To prepare the chilled aqueous extract, 20 grams of Sidr leaf powder were mixed with 400 milliliters of distilled water using an electric mixer. The mixture was allowed to sit at room temperature for 24 hours. It was then filtered through multiple layers of surgical gauze to remove plankton. The filtered mixture was centrifuged at 3000 rpm for ten minutes, followed by further filtration using Whatman No. 0.1 filter paper. The liquid extract was dried in an oven at 40°C to achieve a clear solution, which was then refrigerated until needed (Hernandez et al., 1994).

Preparing the Alcoholic Extract

The alcoholic extract was prepared following the method described by Ladd et al. (1978). Twenty grams of Sidr leaf powder were subjected to Soxhlet extraction using 400 milliliters of 95% ethyl alcohol for 24 hours. The resulting extract was then dried in an oven at 40°C.

Preliminary Phytochemical Screening

Alkaloid Detection

Mayer's Reagent: Prepared by dissolving five grams of potassium iodide and twelve grams of mercuric chloride in one liter of distilled water. When a few milliliters of aqueous or alcoholic extracts were added, the appearance of a white to brown precipitate indicated the presence of alkaloids (Al-Ramahi, 1984; Harbone and Antherden, 2006).

Tannic Acid Detector: Prepared as a 1% solution, which precipitates alkaloids. Adding 1-2 ml of the extract to 5 ml of this reagent

resulted in a white to brown turbidity, confirming the presence of alkaloids (Al-Darwish, 1983).

Phenol Detection

1% Lead Acetate Reagent: An aqueous solution of 1% lead acetate was used to detect tannins. Mixing an equal amount of this reagent with the extract resulted in a bluish-green precipitate, indicating the presence of phenols (Al-Mukhtar, 1994; Al-Salami, 1998).

1% Potassium Hydroxide (KOH): Used to detect furanocoumarins and flavonoids. Adding 1% KOH solution to an equal amount of the extract produced a yellow-green color, indicating the presence of these compounds (Harbone, 1984).

Terpenoid Detection

Foam Reagent: The presence of saponins was detected by shaking a sealed bottle containing the aqueous extract. The appearance of dense foam that persisted indicated the presence of saponins (Harbone, 1984; Al-Mukhtar, 1991).

Mercuric Chloride (HgCl₂) Reagent: Adding 1-2 ml of HgCl₂ to 5 ml of the extract resulted in a white precipitate, indicating the presence of saponins (Al-Mukhtar, 1994; Harbone, 1984).

Preparation of Extract Solutions

For the aqueous extract, solutions with concentrations of 100 mg/ml and 200 mg/ml were prepared. For the alcoholic extract, two grams of dry matter were dissolved in three milliliters of ethyl alcohol, then diluted to ten milliliters with distilled water to achieve a final concentration of 200 mg/ml. Other required concentrations were prepared similarly.

Microbial Specimens

Four bacterial isolates were obtained from the microbiology laboratories at the University of Mosul's College of Sciences. After calibrating the isolates using a McFarland tube, they were spread on Mueller-Hinton medium to assess the effects of the Sidr plant extract. The agar well diffusion method was used, with wells of six millimeters diameter drilled into the medium. Each well received 0.1 milliliters of bacterial cell suspension. Various concentrations of plant extract, as described earlier, were added to the wells. Distilled water served as a negative control. After a 15-minute incubation period, the culture plates were incubated for 24 hours at 37°C. The inhibition zones were measured using a ruler to determine the antimicrobial activity of the extracts.

Results and Discussion

Chemical Composition of Sidr Plant Extracts

The initial chemical analysis of the Sidr plant indicated a rich composition of active ingredients. Notably, alkaloids such as spinanina and compounds derived from jujube fruit were identified, both known for their antibacterial properties (Al-Abed, 2008). Additionally, the Sidr plant contains a variety of other bioactive compounds including phenols, saponins, pectin, lipids, tannic acid, and antioxidant glycosides, along with several types of

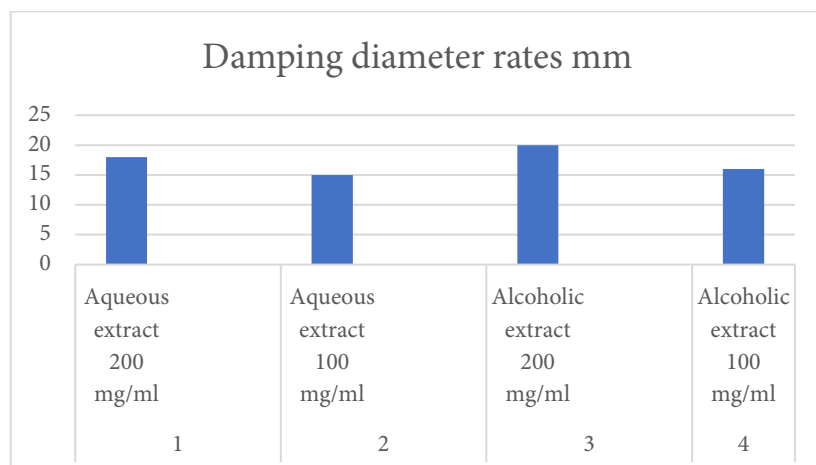


Figure 1. The influence of varying doses of Sidr plant aqueous and alcoholic extracts on *Streptococcus feacalis* thickening's typical diameter.

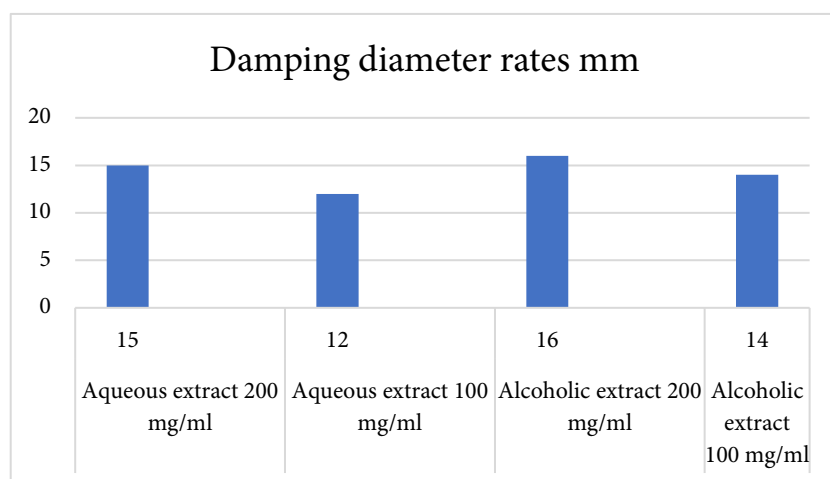


Figure 2. The consequences of varying doses of Sidr plant aqueous and alcoholic extracts on *Staphylococcus aureus* constriction diameter rates.

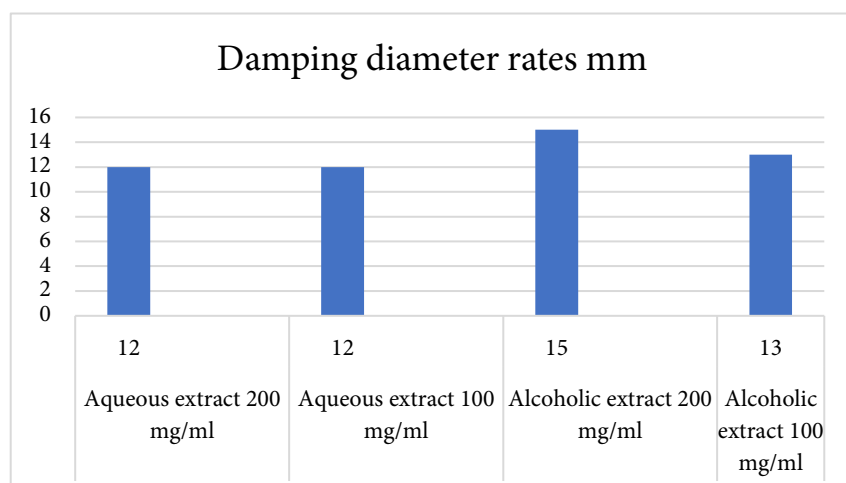


Figure 3. The impact of varying doses of Sidr plant aqueous and alcoholic extracts on *Escherichia coli* inhibition diameter rates.

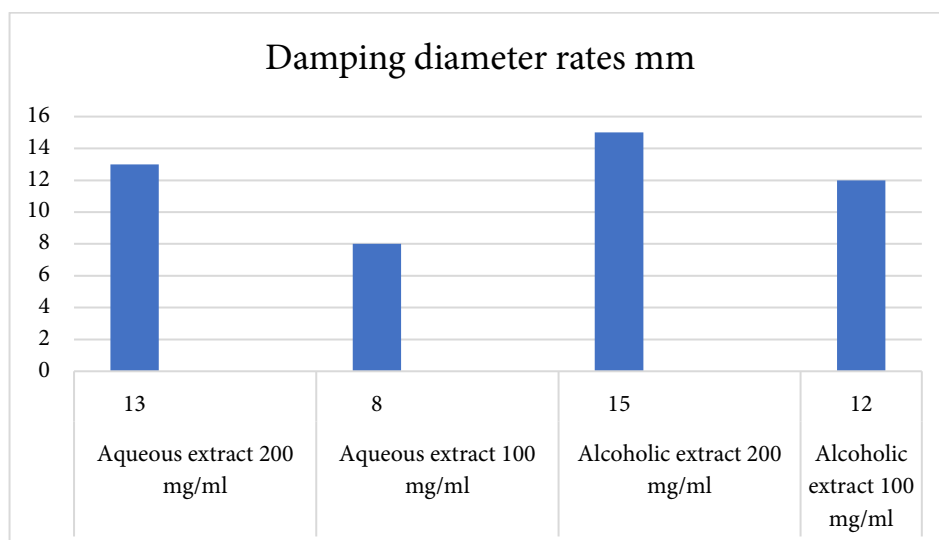


Figure 4. The influence of different amounts of aqueous and alcoholic extracts of the Sidr plant on the rate of inhibition diameters of *Klebsiella sp.*

flavonoids. Terpenes, tannins, and zizyphic acid were also present, aligning with findings from Al-Rawi (1964). The plant contributes xanthine compounds and chlorophyll pigments (Chakravarty, 1976), which may enhance its overall therapeutic profile. The distinct and aromatic fragrance of the Sidr plant can be attributed to its volatile oil content (Felberg, 1994; Ellmore). Furthermore, the plant contains glues, vegetable gels, and mucous substances, which might have various practical applications.

Antibacterial Activity of Sidr Extracts

The study investigated the antibacterial efficacy of both alcoholic and aqueous Sidr extracts against different microbial strains. The results indicated that the extracts' antibacterial activity was concentration-dependent, with higher concentrations yielding stronger inhibitory effects.

Inhibitory Effects on Bacterial Growth

Figures 1 to 4 demonstrate the relationship between extract concentration and microbial inhibition. The data revealed that at a concentration of 200 mg/ml, both alcoholic and aqueous extracts showed the highest antibacterial activity. The alcoholic extract was particularly effective, with a maximum inhibition zone of 20 mm observed against *Streptococcus faecalis*. On the other hand, the least inhibitory effect was noted with the aqueous extract against *Klebsiella* sp., measuring an 8 mm inhibition zone at a concentration of 100 mg/ml.

These findings suggest that the alcoholic extract of Sidr is generally more potent in inhibiting bacterial growth compared to the aqueous extract. This could be due to the better solubility of certain active compounds in alcohol, which enhances their availability and interaction with bacterial cells.

The antibacterial properties of the Sidr plant observed in this study are consistent with previous research. Al-Abed (2008) highlighted the antibacterial properties of alkaloids found in Sidr. The comprehensive chemical profile identified in this study supports the traditional use of Sidr in treating various infections. The presence of phenolic compounds and flavonoids, known for their antimicrobial and antioxidant properties, further corroborates the plant's therapeutic potential.

The results of this study underscore the potential of Sidr extracts as natural antibacterial agents. In the context of increasing antibiotic resistance, the use of plant-based extracts offers a promising alternative for managing bacterial infections. The effective concentrations identified in this study provide a foundation for developing standardized formulations for therapeutic use.

Conclusion

In conclusion, Sidr plant contains a diverse array of bioactive compounds with significant antibacterial properties. Both alcoholic and aqueous extracts demonstrated substantial inhibitory effects on bacterial growth, with the alcoholic extract showing higher efficacy.

These findings support the traditional use of Sidr in herbal medicine and highlight its potential as a natural alternative to synthetic antibiotics. Further research is recommended to isolate specific active compounds, elucidate their mechanisms of action, and assess the safety and efficacy of Sidr extracts in clinical settings.

Author contributions

A.N.A conceptualized, designed, and executed the research. Amal collected and analyzed plant samples, performed microbiological assays, interpreted the data, and wrote and revised the manuscript, ensuring its accuracy and integrity.

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Competing financial interests

The authors have no conflict of interest.

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