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# A Study of active medicinal plant (Artemisia Judaica) against Staphylococcus aureus

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## Abstract

Staphylococcus aureus infection is a rather insidious disease. If tests have shown that Staphylococcus aureus is found on the skin, intestines, ears or nasopharynx, Treatment with pharmaceutical drugs extracted from medicinal plants can be a way to get rid of infection. Plants are considered the greatest source to obtain new antimicrobials.

The present study aimed at evaluating the in vitro antimicrobial activity of methanolic extracts of Artemisia Judaica plant against Staphylococcus aureus Sp. and others pathogenic bacteria, these bacteria are commonly found in hospital-acquired infections.

The methanolic extract of Artemisia Judaica effective against the isolates microorganisms, S. *aureus*, *E. coli*, *P. aeruginosa*, The diameter of zone of inhibition was found to be in the range of 14 – 30 mm against various bacterial strains tested, with maximum diameter against bacteria (S. aureus, 30 mm). The methanolic extract of Artemisia Judaica presented the highest anti-staphylococcus aureus activity and was effective against others bacterial strains tested.

**Keywords:** staphylococcus aureus; medicinal plants; artemisia Judaica; antimicrobial activity

## Introduction

Artemisia Judaica is bushy, perennial herbs as shown in figure [1], aromatic, with woody bases and strong spreading branches, covered by woolly hairs, and made of tubular florets. As other Artemisia species, A.Judaica is found in many traditional preparations to treat inflammation and infections caused by fungi, bacteria, and viruses. This plant is widely used. A. Judaica was described by herbalists as traditional agent to treat coronary artery thrombosis and cardiac infarction. Interestingly, in other traditional medicines of the Arabic region, A.Judaica is also used as treat gastro-intestinal disease, and to enhance eyesight, cardiovascular health and improving of connective tissue, appearance of skin, and the immunity, while promoting decreased risk of atherosclerosis, cancer and arthritis. Furthermore, it is reported that A.Judaica too used as traditional medicine of Desert dwellers in Saudi Arabia and Sinai, Egypt as a herbal remedy with anthelmintic, antibacterial, anti-inflammatory, and analgesic activities. It also relieves snake stings, scorpion bites, ear infections, dysentery, coughing and external wounding [1].

**Figure 1:** *Artemisia Judaica*

**The main Purpose and objectives of the research, Study of the effect of the extract of Artemisia Judaica plant on a different group of pathogenic bacteria, especially Staphylococcus aureus.**

## 1. LITERATURE REVIEW

**Artemisia Judaica– contents and applications**

Artemisia /ˌɑːrtɪˈmiːziə/ [2] is a large, diverse genus of plants belonging to the daisy family Aster**.** Artemisia comprises hardy herbaceous plants and shrubs, which are contains of the useful chemical compounds in their essential oils. Artemisia species growing usually in dry or semiarid habitats. Notable species include A. vulgaris (common mugwort), A. tridentata (big sagebrush), A. annua (sagewort), A. absinthium (wormwood), A. dracunculus (tarragon), and A. abrotanum (southernwood). The leaves of many species are covered with white hairs [3].

Most of these types have strong aromas and bitter tastes from terpenoids and sesquiterpene lactones, which discourage herbivory, and may have had a selective advantage, The small flowers are wind-pollinated.[3] Artemisia species are used as food plants by the larvae of a number of Lepidoptera species.

### 1.1.1. Botanical characteristics of Artemisia Judaica

Artemisia Judaica is a herp shrub with yellow flowers as shown in figure (2) and pubescent leaves, which grows widely in the southern desert of Algeria, in Egypt (in Sinai desert), and in the Middle East (Israel, Jordan and Saudi Arabia).[4] It is widely used in Algerian traditional medicine as a vermifuge, stomachic, sedative, diarrheic, analeptic and antispasmodic agent [5].

**Figure 2:** *Artemisia Judaica with yellow flowers*

### 1.1.2. Active phytochemical ingredients of Artemisia Judaica

Chemical analysis of these plants showed that they are rich in flavonoid sources [6] and sesquiterpenes lactones [7,8], A number of volatiles chemical constituents from the aerial parts of A. judaica has been identified; The main compounds are the derivatives of piperitone and cinnamon [9,10].

### 1.1.3. Biological activity of Artemisia judaica and its extraction

The studies on biological activity reported that the extracts of A. judaica showed acaricidal activity [11], allelopatic and antioxidant activities [12]. The essential oil showed insecticidal [13], antifeedant and antifungal properties [14], the aqueous extract of dried leaves of A. judaica grown in the Saudi Arabian desert revealed significant antioxidant effects [15,16]. However, to our knowledge, the effect of the essential oil of aerial parts of A. judaica against multi drug resistant (MDR) pathogenic bacteria isolated from patients had not been reported and nobody has discussed whether processing influenced the medicinal effect of this oil.

### 1.1.4. Pharmacological properties of Artemisia judaica

A. judaica is widely used in traditional medicine being recommended by aboriginal Bedouins in the North Badia region of Jordan as calmative. Furthermore, it is used for the treatment of stomach ache, heart diseases, sexual weakness, diabetes, gastro-intestinal disorders and external wounding. Additionally, other folk medicines of the Arabic region commonly use this aromatic plant for the treatment of inflammatory-related diseases, for instance fungal infections, diabetes, atherosclerosis, cancer and arthritis [1].

### 1.2. The concept of staphylococcus

### 1.2.1. The most common staphylococcal infections are:

Skin infections that often cause abscesses as shown in figure (3) However, bacteria can travel through the bloodstream (called bacteremia) and infect almost any area of the body, especially heart valves (endocarditis) and bones (osteomyelitis). Bacteria also tend to accumulate on medical devices in the body, such as artificial heart valves or joints, pacemakers, and catheters inserted through the skin into blood vessels [10].

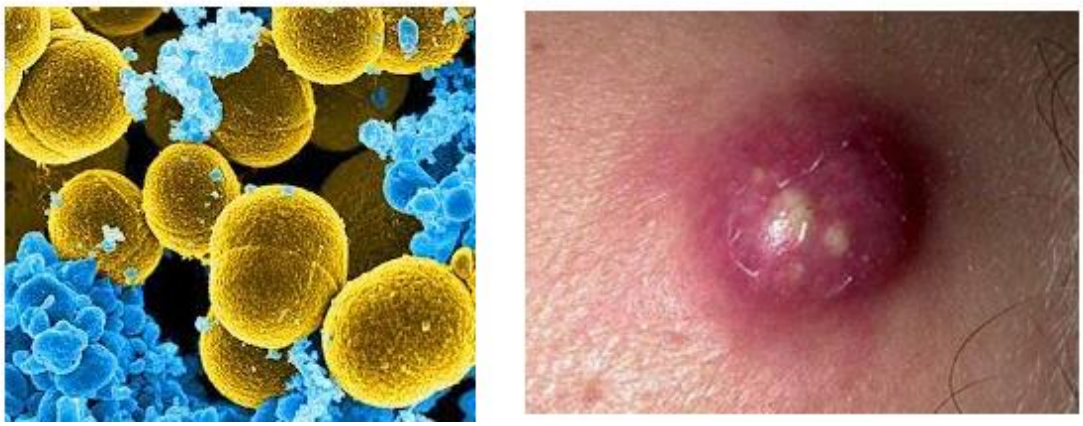

**Figure 3:** *Some staphylococcal infections are more likely in certain situations*

Bloodstream infections: When a catheter, inserted into a vein, remains in place for a long time. Endocarditis: When people inject illegal drugs or have an artificial heart valve, or when a catheter inserted into a vein becomes infected. Osteomyelitis: When Staphylococcus aureus spreads to the bone due to an infection in the bloodstream or an infection in nearby soft tissues, which can occur in people with deep pressure sores or foot ulcers due to diabetes. Lung infection (pneumonia): When people have had the flu (especially) or a bloodstream infection, when people are taking corticosteroids or drugs that suppress the immune system (immunosuppressants), or when they are hospitalized for tracheal intubation and mechanical ventilation (called hospital pneumonia) Staphylococcus toxins Staphylococcus toxins There are many strains of Staphylococcus aureus. Some strains produce toxins that can cause staphylococcal food poisoning, toxic shock syndrome, or skin burn syndrome. Toxic shock syndrome is also caused by toxins produced by some streptococci. This syndrome causes rapidly progressive and severe symptoms, including fever, rashes, dangerously low blood pressure, and multiple organ failure [17].

In the absence of timely and adequate treatment of infection caused by S. aureus, severe complications develop:

- **Sepsis,**
- **Meningitis,**
- **Endocarditis,**
- **Infectious toxic shock,**
- **Coma,**
- **Fatal outcome.**

The prognosis of the disease is ambiguous. It is determined by the severity of the pathology. Mild forms involving skin and mucous membranes in the pathological process are completely cured without negative consequences. Sepsis, brain damage and other serious complications often end in death [18].

### 1.2.2. Measures to avoid the development of staphylococcal infection:

1. Strengthening immunity - hardening, sports, proper nutrition, good sleep, walking in the fresh air,

2. Timely treatment of infectious diseases and sanitation of foci of infection - caries, tonsillitis, urethritis,

3. Compliance with the rules of hygiene - frequent hand washing, wet cleaning in the room, high-quality food preparation,

4. Restricted visits to public places during the peak of respiratory diseases.

Preventive measures on a national scale include constant monitoring of the sanitary and epidemiological regime in health care facilities, routine examination of medical workers in maternity hospitals and surgical departments, timely detection of carriers of staphylococcus aureus, immunization of persons at risk with toxoid or immunoglobulin. Diseases caused by Staphylococcus aureus progress rapidly without appropriate treatment. At the same time, the inflammatory process easily passes from the primary focus to neighboring organs and tissues. In a weakened body, infection of any organ can occur. Self-medication and unwillingness to seek medical help from a doctor usually ends in complications and even death of patients [18].

Biofilm production is an important contributor to the virulence of microorganisms associated with chronic infections such as sinusitis, otitis media, cholecystitis, prostatitis, osteomyelitis, chronic skin infections and infections associated with foreign bodies (implants and catheters), with S. aureus often identified as important the agent responsible for such infections [19]. Both S. aureus and Staphylococcus epidermidis are the most clinically significant pathogens among Gram-positive bacteria capable of forming biofilms [20].

The ability of these bacteria to adhere to abiotic surfaces of medical devices and form stable biofilms on them contributes to the pathogenicity of staphylococcal infections [21]. After the installation of a medical device, its polymer-based material quickly becomes covered with plasma proteins and extracellular matrix, thereby enhancing the microbial colonization of its surfaces [22]. In addition to indirect binding of bacteria to the polymer, direct nonspecific binding can also occur, mainly due to electrostatic and hydrophobic interactions promoted by bacterial surface proteins [23].

Curcumin is a polyphenolic compound isolated from crushed rhizomes of the Curcuma longa L. plant [24]. This plant has a wide spectrum of biological effects [25, 26], including antioxidant, analgesic, anti-inflammatory, antiseptic, antitumor, antiviral, antibacterial, antifungal and antiplatelet effects [27, 28]. Curcumin has been widely used in Ayurvedic medicine for centuries, and there have been no reports of toxicity [27, 28]. Research over the past 50 years has shown that polyphenols such as curcumin play an important role in maintaining health and preventing disease [26]. A number of studies have shown that C. longa extract and fractions have antibacterial activity against pathogenic bacteria, including S. aureus [29, 30]. The study showed that the MIC of curcumin, the main compound isolated from C. Longa, ranged from 125 to 250 μg ml $^{-1}$ versus ten methicillin-susceptible Staphylococcus aureus (MSSA) and MRSA strains. In addition, a combinatorial checkerboard test showed that curcumin reduces the MIC of antibiotics commonly used against MRSA, such as oxacillin, ampicillin, ciprofloxacin, and norfloxacin by 2-128 times, thus demonstrating the potential clinical efficacy of curcumin and its derivatives for the treatment of MRSA infection [ nineteen]. Many other authors have also studied the effect of curcumin on fungal pathogens [31, 32].

## 2. Materials and Methodology of the experiment

### 2.1. Plant extraction

The collected plant samples were dried under shade and powdered mechanically using a commercial electrical stainless-steel blender and extracted by soaking in 80 % Methanol for about five days with occasional shaking. The solvent was evaporated under reduced pressure to dryness using rotary evaporator apparatus (Rotary evaporator Büchi R-3000 (Büchi, Switzerland), and the extract allowed to air till complete dryness. The extract were then stores in refrigerator until usea as shown in figure [4].

**Figure 4:** *Artemisia Judaica Extract*

#### 1.3.2. Test microorganisms

The extract was tested against seven reference strain bacteria *Staphylococcus aureus*(ATCC 254996),*Enterococusfecalis (ATCC* 254602*) Streptococcus pneumoniae*(ATCC 254657), *Escherichia coli* (ATCC 254607*), Proteus mirabilis (ATCC 257440),Pseudomonas aeruginosa* (ATCC 254992*), Klebsiella pneumoniae (*ATCC 254656)and one standard fungus, *Candida albicans*(ATCC 254625).[33] The tested organisms were obtained from the Department of Microbiology, King Fahad Hospital, Jazan, Saudi Arabia

#### 1.3.3. Preparation of the test organisms

The turbidity of growing bacteria was adjusted and matched with the turbidity of 0.5 McFarland units by the inoculation of the tested bacteria in 4ml of peptone water and allowed to incubate for hours at 37 °C [34].

The candida albican was cultured on Sabouraud dextrose agar mediumand then incubated for four days at 25 °C. The candida suspension was harvested and wash with 100 ml sterile normal saline and store at 4 °C for further used [34].

### 2.2. Antimicrobial susceptibility for the plant extract

Antimicrobial efficacy test of the plant extract was accomplished by using agar diffusion method with minor modification [35], One ml of standardized stock suspension containing 105 - 106 C.F.U/ mL was mixed with 100 ml of Muller Hinton agar medium and then kept at 45 °C. The media were then distributed into sterile plates and kept to set. Holes were then made by using sterile cork borer number 4. The holes were filled with 50µl of plant extract and allowed to get diffused at room temperature for minimum 1hour. Theplates were kept in incubator over night at 37 °C temperature. The diameters of resulting inhibition zone were then measured. Similar procedure was done for testing antifungal activity. Sabouraud-dextrose agar medium was used instead of Muller Hinton agar, then incubated for 2 days at 25 °C [35].

### 2.3. Minimum inhibitory concentration (MIC)

Detection of the minimum inhibitory concentration (MIC) was adopted for the crude extract of (extract *Artemisia Judaica*) against the microorganisms using broth dilution method. Test bacterial cultures (100 μL of bacterial culture containing 105 CFU/mL) were inoculated into tubes containing different concentrations of extract of 25, 12.5, 6.25, 3.12,1.56, 0.78 and 0.39 mg/L and incubated overnight at 37 °C. The values were determined by detecting the inhibition of visible growth in the culture tubes. Similarly, Minimum Bactericidal Concentration (MBC) was detected by sub- culturing the broth onto freshly Muller Hinton agar medium and at 37°Cover night. The last concentration of MIC tubes which didn't Shaw any growth of bacterial was regarded as MBC [36].

## Results and Discussion

**Antibacterial efficacy: -**

The antimicrobial efficacy of the plant extract was examined by the disc-diffusion method and the results are shown in (Table (1), figure5a,5b,5c). Plant extract (extract *Artemisia Judaica*) was found to be effective against the isolates microorganisms, S. *aureus, E. coli, P. aeruginosa,* The diameter of zone of inhibition was found to be in the range of 14 – 30 mm against various bacterial strains tested, with maximum diameter against bacteria (S. aureus, 30 mm). Antibacterial activity measured by the disc-diffusion method has some limitations as it only indicates the bacterial growth inhibition without any evidence of the bacteriostatic or bactericidal action of the extract. Therefore, determination of MIC and MBC values of extract is used to establish the dose specificity and nature of the activity of the extract. Minimum inhibitory concentration (MIC) values of plant extract was done by estimated using brothmacro-dilution method, whereas, MBC values were determined by sub-culturing all prepared concentrations (≥ MIC) with no detectable growth. The MIC and MBC values of the plant extracts were calculated to be in the range of (15.6 and 250 mg/L, respectively against all bacterial strains tested as shown in Table (1).

| **Organism** | **Plant extract** | | | **Control Amikacin** |
|---|---|---|---|---|
| | **Zone inhibition** | **MIC** | **MBC** | |
| *E. coli* | 20 | 250 | 500 | 35 |
| K. pneumonia | - | - | - | - |
| *P. mirabilis* | - | - | - | - |
| *P. aeruginosa* | 14 | 250 | 500 | 30 |
| *Streptococcus pneumoniae* | - | - | - | - |
| S. *aureus* | 30 | 15.6 | 31.25 | 34 |
| Enterococusfecalis | - | - | - | - |
| Candida albican | - | - | - | |

**Table 1:** *Inhibition zone, minimum inhibitory concentration (MIC) and minimum bactericidal concentration (MBC)*

**Figure (5a):** *Antimicrobial efficacy of (Artemisia Judaica) against E.coli*

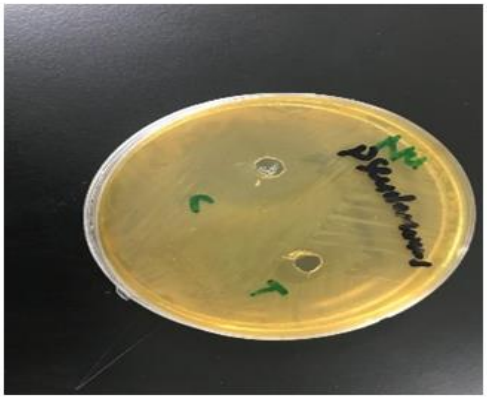

**Figure (5b):** *Antimicrobial efficacy of (Artemisia Judaica) against P. aeruginosa*


**Figure (5c):** *Antimicrobial efficacy of (Artemisia Judaica) S. aureus*

## Conclusions

The extract was tested against seven reference strain bacteria *and one fungus*, The extract was active with only Three of them. The study demonstrated in vitro that the methanolic extract of the plant had a high activity against Staphylococcus aureus bacteria.

**Conflict of Interest:** No conflict of interest.

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