Abstract: Three bacterial has been isolated from pus of wound foot located at the Saudi Hospital, at Hajjah-Yemen such as Pseudomonas sp, Klebsilla sp, and Provotella sp. Antibacterial efficacy was elucidated using disc diffusion method. The alcohol extract of leaf and stem showing highest inhibitory activity on all the four bacterial species tested by using extract between 0.005-0.02 mg/disc.

2. Materials and Methods

2.1 Source of microorganisms

Bacteria strains were isolated from samples collected from pus of wound foot located at the Saudi Hospital, at Hajjah-Yemen (Plate1 (a) and (b)). All colonies that had grown were identified according to the descriptions in the Bergeys Manual of Systematic Bacteriology, Vol.1 (1984), Vol.2 (1986).

2.2 Collection of plant material

The fresh yellow latex and extract of leaf and stem for study include leaf and stem of Argemone mexicana. This plant were collected from the Yemen Mountain is used as medicine wildly and identified by a botanist [Plate 2 (c) and (d)]. The fresh yellow latex was collected from the leaf and stem and filter with a pore size diameter of 0.45 µm and then stored until further use. The fresh picked parts of the plants were rinsed with running tap water and then with distilled water. They were air dried at room temperature for 2 weeks, with no direct sunlight. Once dried, plant materials were ground and stored at 4°C before subjecting them individually to solvent extraction procedures this is method according to [5].

2.3 Preparation of crude extracts

The method of solvent extraction consisted of soaking the ground plant materials in Alcohol solvent for 24h, followed by shaking in an incubator shaker set at 200 rpm and 35°C for 4h. The standard plant material weight/solvent volume concentration (w/v) was used 1/10. The extracts were filtered using Whiteman filter paper no.1. The supernatant was taken and concentrated on a hotplate set at 38°C until the solvents evaporate completely. The obtained solvent extracts were weighed and stored at 4°C until further use [5] method.

2.4 Antibacterial assay by disc diffusion method

Antibacterial activity was demonstrated using single disc diffusion method [1]. A pure colony of each the test organisms were sub-cultured into a 5 ml of Normal saline solution was prepared by dissolving 8.5g NaCl in 1 L distilled water and then autoclaved at 121 °C for 15 mins, followed by incubation at 37°C for 24h. The test was carried out by placing each disc (5mm) impregnated with 0.03mg of the extracts disc on Nutrient Agar (NA) surface previously inoculated with 100μl suspension of test organism. Respective solvent without plant extracts served as negative control. Standard antibiotic disc of Ciprofloxaxine was used (0.03 mg/disc) as positive control. Plates were incubated at 37°C for 24h to observe formation of clear zone of inhibition.
3. Results and Discussion

**Identification of bacteria:** Biochemical and characteristics of three isolates (Pseudomonas sp, Klebsilla sp, and Provotella sp) exhibited good potential were further investigated. The results of the fresh yellow latex and extract of leaf and stem for study of Argemone Mexicana and results of MIC assay of fresh yellow latex, distilled water extract of the leaf and stem and alcohol extract of leaf and stem Table 1. The MIC impregnated with different concentration of the extracts ranging from 0.005-0.04 mg/disc were tested against the organisms Table 1. Distilled water extract of the leaf showed highest inhibitory of growth all of three isolated Pseudomonas sp, Klebsilla sp, and Provotella sp respectively further aqueous extract of the stem showed near to highest inhibitory of growth of Pseudomonas sp, Klebsilla sp, and Provotella sp respectively. [5] also reported that distilled water extract of the leaf showed highest or near to highest inhibitory activity on all the bacterial species tested during the study. The second highest inhibition of growth was showed with fresh yellow latex of Pseudomonas sp, and Klebsilla sp compared to distilled water extract of leaf and stem father growth inhibitor activity showed relatively less of inhibitory effect of Provotella sp with fresh yellow latex. [4] reported that the fresh yellow latex contains protein-dissolving substances effective in the treatment of warts, sores and skin infection. It is also used in curing dropsy and jaundice. The alcohol extract of leaf and stem showed relatively less of inhibitory effect on growth of the test isolated. (Bhattacharjee et al., 2006) report on growth inhibitory effect of solvent extracts of A.mexicana on few other species of pathogenic bacteria.

The effect of activities of the aqueous extract of leaf and also activity of fresh yellow latex on Pseudomonas sp with different concentration of the extracts ranging from 0.005-0.03 mg/disc showed in (Plate 3and 4. Our study exhibited good potential were further investigated of the fresh yellow latex and extract of leaf and stem for study of Argemone Mexicana current result has supplemented earlier report that the antibacterial compounds from plants may inhabit bacterial growth and may be of significant clinical value in treatment of patients infected with resistant strains.

**Table 1:** Minimal inhibition concentration (MIC) in mg/disc of the selected extracts of A.mexicana and solvent extracts.

<table>
<thead>
<tr>
<th>Organism</th>
<th>Extract of leaves</th>
<th>Extract of stem</th>
<th>Control</th>
</tr>
</thead>
<tbody>
<tr>
<td>Pseudomonas sp</td>
<td>0.04 D.W, 0.005 Alcohol</td>
<td>0.005 D.W, 0.01 Alcohol</td>
<td>0.005 mm Ciprofloxaine</td>
</tr>
<tr>
<td>Klebsilla sp</td>
<td>0.02 D.W, 0.04 Alcohol</td>
<td>0.03 D.W, 0.01 Alcohol</td>
<td>0.005 mm Ciprofloxaine</td>
</tr>
<tr>
<td>Provotella sp</td>
<td>0.01 D.W, 0.03 Alcohol</td>
<td>0.02 D.W, 0.01 Alcohol</td>
<td>0.005 mm Ciprofloxaine</td>
</tr>
</tbody>
</table>

![Figure 1: (a) and (b) - Samples from the Saudi Hospital, at Hajjah-Yemen](image1.png)

![Figure 2: (c) and (d) - Argemone Mexicana Linn collected from Yemen Mountain](image2.png)
Figure 3: MIC assay of aqueous extract of leaves on *Pseudomonas sp*

Figure 4: MIC assay of fresh yellow latex on *Pseudomonas sp*

References


